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## Vitality and Germination of Seeds.

A THESIS PRESENTED FOR THE DEGREE OF

DOCTOR OF SCIENCE AT THE UNIVERSITY

OF MICHIGAN IN JUNE, 1902.

BY

J. W. T. DUVEL,

HOLDER OF THE DEXTER M. FERRY

FELLOWSHIP IN BOTANY.



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### THE VITALITY AND GERMINATION OF SEEDS

#### INTRODUCTION.

It has long been known that the conditions under which plants are grown and the degree of maturity at the time of harvesting are factors which play an important part in the life of seeds. But, granting that seeds are of strong vitality at the time of harvesting, there remain to be considered the methods of gathering and curing, the water content of the seed at the time of storing, the methods of storage, the humidity and temperature of the surrounding atmosphere, the composition of the seed, the nature of the seed coats, activities within the cells, and numerous other factors which play important parts in the life of the seed.

The conditions necessary for the successful germination of a seed of good vitality and the chemical transformations accompanying these early stages of development have received considerable attention from numerous investigators. These changes and conditions are fairly well understood for many of our common seeds. However, several important facts still remain unexplained, and our knowledge will not be complete until each and every species has been carefully studied.

On the other hand, the conditions influencing the vitality of seeds as commercially handled are but little understood and have been almost wholly neglected in research work. Likewise, but little attention has been given to the complex chemical and physical changes which take place in mature seed during the slow process of devitalization. It was in order to determine some of these factors that the work described in these pages was begun, and the results are thus of considerable practical value as well as of scientific importance. The present paper treats chiefly of the conditions influencing the vitality and germination of seeds when subjected to such methods of treatment as are generally met with in the ordinary handling of seed. Particular attention has been given to the effect of climate, moisture, and temperature on vitality, supplemented with a discussion of the changes taking place in mature seeds, especially the respiratory activities and the part played by enzymes.

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The results of the above experiments have suggested improved methods of storing and shipping seeds so as to prolong their vitality and also to secure the production of more vigorous seedlings.

The work for the present paper was begun in 1899 at the University of Michigan and was continued for three consecutive years while the writer held the Dexter M. Ferry Botanical Fellowship in that institution. During this time the investigation was under the direction of Prof. V. M. Spalding, Ph.D., and Dr. F. C. Newcombe, who showed great interest in it and gave valuable suggestions as the work progressed, at the same time placing the facilities of the laboratory and of the library at the disposal of the writer. Since September 1, 1902, the work has been continued in the Seed Laboratory of the U. S. Department of Agriculture. Valuable assistance in storing seeds was rendered by Prof. C. W. Burkett, at Durham, N. H.; Mr. E. E. Smith, Wagoner, Ind. T.; Prof. W. R. Dodson, Baton Rouge, La.; Prof. F. S. Earle, Auburn, Ala.; Zimmer Brothers, Mobile, Ala.; Prof. H. H. Hume, Lake City, Fla., and Prof. Charles B. Scott, San Juan, Porto Rico.

#### MATERIALS AND METHODS.

#### SEEDS.

For these experiments thirteen different samples of seeds were used, being so selected as to include representatives of ten different families and twelve genera and species, as follows:

Poacex—Zea mays, sweet corn (two samples).

Liliaceæ—Allium cepa L., onion.

Brassicaceæ—Brassica oleracea L., cabbage; Raphanus sativus L., radish.

Apiaceæ—Daucus carota L., carrot.

Fabaceæ—Pisum sativum L., pea; Phaseolus vulgaris L., bean.

Violaceæ-Viola tricolor L., pansy.

Polemoniaceæ—Phlox drummondii Hook, phlox.

 $Solanace \&-Lycopersic con\ lycopersic cum\ (L.)\ Karst.,\ to mato.$ 

Cucurbitaceæ—Citrullus citrullus (L.) Karst., watermelon.

Asteraceæ—Lactuca sativa L., lettuce.

It will thus be seen that the seeds used cover a wide range as to family characteristics, as well as size, structure, and composition of seed. Likewise they are all from plants of the garden or field that have undergone a high degree of cultivation, thus enabling the seeds to withstand more or less variation as to conditions of vitality and growth.

All seeds used throughout these experiments were provided by D. M. Ferry & Co., of Detroit, Mich., and the seed furnished was of strong vitality and of known age and origin. The corn "A" (Minnesota Sweet), onion (Yellow Danvers), pea (D. M. Ferry Extra Early), bean (Yellow Kidney, Six Weeks), tomato (Dwarf Champion), and the

watermelon (Sweet Mountain) were grown in Michigan. The corn "B" (Minnesota Sweet), was grown in Nebraska, the cabbage (Winningstedt), in Washington, and the lettuce (Black-Seeded Simpson), in California, while the radish (Early Scarlet Turnip-Rooted), carrot (Chantenay), pansy (mixed), and Phlox drummondii (mixed) were grown in France. The seed was all of the harvest of 1899 and was received at the botanical laboratory of the University of Michigan on January 27, 1900.

On January 30, 1900, germination tests were made, showing the vitality of the seeds to be as follows:

Kind of seed,	Percentage of germination.	Kind of seed.	Percentage of germination.
Bean	100	Pansy	69.5
Cabbage	93	Pea	97
Carrot	83.5	Phlox	78
Corn, sweet, "A"	94	Radish	81
Corn, sweet, "B"	88 .	Tomato	98
Lettuce	87.5	Watermelon	99

Vitality of seeds tested January 30, 1900.

#### GERMINATION TESTS AND APPARATUS.

In the preliminary work several methods of testing were tried, but as none proved as serviceable as the "Geneva tester," this apparatus was adopted for all subsequent tests as recorded in the following pages. The detailed construction of this tester need not be described, for it is simple and quite familiar to all. However, some modifications were made in the preparation of the apparatus, and some precautions taken in the manipulation, which have proved to be of much value. The brass wires originally and ordinarily used to support the folds of cloth were replaced by glass rods of 6 to 7 mm. diameter. Rods of this size are much heavier than is necessary to support the folds of cloth, but the chief advantage in having rods of large diameter is that in case of the germination of large seeds the folds can be drawn near together at the top and still have sufficient space within the fold for the seeds. On the other hand, in the germination of small seeds that require considerable quantities of air, the folds can be closed at the top by bringing the rods together, thus insuring more uniform conditions throughout the fold and at the same time leaving sufficient space above the seeds for an abundant supply of air. The chief advantage in substituting glass rods for brass wires is in removing the possible source of injury resulting from the poisonous action of the dissolved copper.

Another error frequently, if not always, made in using such a tester is in allowing the ends of the cloths, or sometimes the bottoms of the folds, to dip into water in the pan. This should never be permitted, for in that way seeds are kept too moist, especially near the ends of the folds. Likewise such methods give an opportunity for the transmission of dissolved copper and a resulting injury to the seeds. For this same reason the strips of cloth should be made sufficiently narrow not to come into contact with the sides of the pan.

Much better results are obtained if the seeds, before being placed in the germinator, are soaked in water for several hours, the length of time depending on the power of absorption of the seeds. In these experiments the seeds were always soaked in distilled water for twelve or fifteen hours before transferring them to the germinator. This preliminary soaking gives a more speedy germination, which is always advantageous, especially in making comparative germination tests. In order to supply the requisite amount of moisture for subsequent growth, the cloths were first uniformly and completely wet with distilled water; moreover much care was taken to see that there was only a very small quantity of water in the bottom of the pan. In case of seeds that germinate readily, such as cabbage, lettuce, and onion, it is necessary that all surface water be removed from the bottom of the germinator if good results are desired. The pan then being covered with a glass plate, it is seldom necessary to increase the amount of moisture, for seeds when once soaked need only to be kept slightly moist and not wet, as must necessarily be true if the ends of the cloths or bottoms of the folds dip into the water. After soaking, the water in the seeds and cloths is ample for the completion of most germination tests. However, in an occasional test the seeds may become slightly dry, which happens when the cover is kept off the pan for a considerable time while counting germinated seeds. In such cases the remedy is to pour a small quantity of water in the bottom of the pan. or in extreme cases to moisten the folds with a fine spray.

If the above modifications be adopted and the necessary precautions taken, many of the objections frequently made to the Geneva tester will be removed and the difficulties will be overcome; at least it is a most excellent method of testing seeds where comparative results are especially desired. It must also be borne in mind that the Canton flannel (which is generally used in making the pockets) should always be of the best grade and should never be used a second time without being thoroughly cleaned and sterilized.

In selecting samples for germination the impurities and the immature seeds were first removed. The samples for test were then made up of the remaining large and small seed. For the most part 200 seeds were taken for a test, but with the larger seeds—corn, pea, bean, and watermelon—100 seeds were usually used. In all cases where any irregularity was apparent, tests were repeated. The controls are based on the results of several duplicate tests.

All germination tests were made in a dark room where the temperature could be comparatively well regulated and was maintained nearly constant throughout most tests. Germinated seeds were removed daily during early stages of the tests and a complete record of the number germinating each day was kept. This is of value in seed testing, because the germinative energy of a seed tells much as to its vitality. If seeds have a high vitality, the germinative energy will be very strong, i. e., germination will take place rapidly, giving rise to strong and vigorous seedlings; but if the seeds are of very low vitality, there will be a corresponding retardation in germination, giving rise to weak seedlings, i. e., showing a low germinative energy. In most cases throughout this work only the final percentages of germination are tabulated.

#### EFFECT OF CLIMATIC CONDITIONS ON THE VITALITY OF SEEDS.

It has long since been known that seeds under ordinary conditions lose their power of germination after the lapse of a few years, or in some cases within a few weeks or months. Many investigators have also learned that the rapidity with which seeds lose their vitality, when stored under ordinary conditions, varies greatly with the section of the country in which such seeds are kept. This loss in vitality is especially marked in the case of seeds stored in places of relatively high humidity. The rapid deterioration of seeds in localities having a humid atmosphere has become a source of much embarrassment to seedsmen, for they have experienced many difficulties in shipping seed to such places. This is especially marked in the case of seeds sent to growers or dealers in the vicinity of the Gulf of Mexico. Gardeners and planters in that part of the United States are continually complaining about the nonviable seeds sent out by seedsmen. Some growers have learned how to guard against this difficulty to a certain extent. Zimmer Brothers, of Mobile, Ala., wrote, on February 28, 1900, concerning this matter, as follows:

During thirty years' experience in market gardening, we have learned that seeds of many hardy plants will not keep in our climate, and when ordering we so time our order that we can plant the seeds as soon as received. If such be impossible, we are very careful to keep the original package unopened until conditions are favorable for planting. If we find it necessary to keep seeds of hardy plants for some months, we put them up on arrival in dry bottles, put on top a bit of cotton saturated with chloroform and cork tightly. We have kept, in that way, cauliflower seed satisfactorily for twelve months. At the shore seeds keep very badly; one-half mile back they do much better. As a rule seeds of tender plants give but little trouble.

As far as has been ascertained, no definite experiments have been made with these points in view, and especially with the idea of determining the cause or causes of this deterioration of vital energy. In order to obtain reliable data on these points, a series of experiments was undertaken in February, 1900, to determine how seeds are affected

when distributed to different parts of the United States and submitted to the free influence of various climates. Likewise at the various points where tests were made the seeds were subjected to different treatments.

The places selected for these tests were San Juan, P. R., Lake City, Fla., Mobile, Ala., Auburn, Ala., Baton Rouge, La., Wagoner, Ind. T., Durham, N. H., and Ann Arbor, Mich.

A sample of each species of seed was put up separately in double manila coin envelopes and in closely corked bottles. Duplicate sets of each series were then subjected at each of the above-named places to the following conditions:

Trade conditions.—Conditions similar to those in which seeds are kept when offered for sale by retail dealers, the seed being more or less exposed to meteorological changes and subjected to natural variations in temperature and humidity. For the most part the seeds were in rooms that were never heated.

Dry rooms.—Rooms in the interior of buildings which were artificially heated during cold weather, and where the quantity of moisture was relatively small and the temperature comparatively constant.

Basements.—Rooms where the temperature was comparatively low and uniform, and the relative humidity of the surrounding air was much higher than in "trade conditions" and "dry rooms."

These conditions varied in the different places at which tests were made, and a more detailed description will be given when the results of the germination tests are discussed.

For the first part of this paper, treating of the influence of climate on vitality, none of the seeds need to be considered save those prepared in paper packages and kept under trade conditions, these coming more nearly under the direct action of the surrounding atmosphere. A sample of each kind of seed was put up in a manila (No. 2) coin envelope, and each of these packages was then inserted in a second (No. 3) coin envelope. Duplicate samples of every kind of seed were sent to the various testing places, where they were subjected to trade conditions. At San Juan the packages of seeds were kept in an open room, being subjected to the full action of the atmosphere but protected from the direct rays of the sun and from rain. At Lake City the packages were kept in a one-story frame building which was not artificially heated and the doors of which were open the greater portion of the time. At Mobile the packages of seeds were stored in a comparatively open attic of a private dwelling. At Auburn the seeds were stored in a greenhouse office, with the doors frequently standing open. At Baton Rouge the packages were kept on a shelf in a grocery store, the doors of which were closed only during the night. At Wagoner the conditions were very similar to those of Baton Rouge. save that the packages of seeds were kept in a drug store. At Durham the seeds were kept over a door at the entrance of one of the college buildings. This door opens into a hall which communicates with the offices, chemical laboratory, and the basement. At Ann Arbor the seeds were stored in the botanical laboratory, with slightly varying conditions, they being near a window which was frequently open during the summer, and at irregular intervals during the early part of the summer the packages were placed in the window so as to receive the direct rays of the sun. The seeds stored at Ann Arbor served partially as controls for those sent to the various other places, and, in addition to the last-named series, seeds from the original packages, as received from D. M. Ferry & Co., were kept in a dry and comparatively cool closet on the fourth floor of the botanical laboratory. These seeds served as checks for the complete set of experiments, and are designated throughout this paper as "Control."

The samples were sent out to the above-named places in February, 1900. The first complete set was returned in June, or early July, of that year. The second complete set was allowed to remain throughout the entire summer, and was returned in October and early November of the same year. The average time of treatment for the two series of experiments was 128 and 251 days respectively. When the seeds were returned, germination tests were made as soon as possible. The length of time that the seeds were in the various places and the vitality as shown by the germination tests are given in Tables I and II. In both tables the columns from left to right, beginning with Mobile, Ala., are in the order of the degree to which the seeds were injured.

Table I.—Effect of climate on vitality, as shown by percentage of germination—first test.

Kind of seed.	Con- trol.	Mobile, Ala., Feb. 17 to July 7. 140 days.	San Juan, P. R., Feb. 9 to June 20. 129 days.	Baton Rouge, La., Feb. 17 to June 18. 121 days.	Wagon- er, Ind. T., Feb. 17 to June 23. 126 days.	Lake City, Fla., Feb. 9 to June 18. 129 days.	Durham, N. H., Feb. 17 to July 14. 147 days.	Auburn, Ala., Feb. 17 to May 30. 102 days.	Ann Arbor, Mich.
Corn, sweet, "A"	95, 9	80.0	96. 0	96.0	96.0	94.0	100.0	96.0	100.0
Corn, sweet, "B"	89.3	48.0	72.0	80.0	70.0	86.0	89.3	88.0	92.0
Onion	95.8	7.0	84.5	90.0	93.5	95.0	96.5	96.0	95.0
Cabbage	92.7	64.5	82.0	88.5	83.5	89.5	93.0	91.0	96.0
Radish	83.6	58.5	64.0	77.5	77.5	79.0	80.6	75.5	82.5
Carrot	83.3	59.0	71.5	74.3	81.5	76.5	78, 0	84.5	76.0
Pea	95.3	69.2	94.0	94.0	98.0	96.0	98, 0	93.3	90,0
Bean	98.7	58.0	100.0	96, 0	96.0	98.0	100.0	98.0	98.0
Pansy	63.0	3.0	20.0	28, 5	48.5	44.5	55.5	57.5	53.5
Phlox drummondii	69.0	0.5	23.5	47.5	50.5	41,5	67.0	61.5	67.0
Tomato	95, 5	90.0	94.0	91.5	96.5	91.0	94.5	95.0	89.0
Watermelon	98, 3	98.0	96.0	100.0	98.0	98.0	98.0	94.0	100.0
Lettuce	81, 6	63.0	79.0	82.5	78.0	87.0	82.0	86.5	82.0
Average of all seeds.	87.79	53.59	75.12	80.48	82.12	83.00	85.57	85.70	86.23

From Table I it will be seen that the loss of vitality in the case of seeds stored at Mobile was much greater than in those stored at any of the other places. The greatest loss in the samples tested was in the

phlox, where the germination was only 0.5 per cent, or a loss in vitality of 99.3 per cent as compared with the control. These results were closely followed by a loss in vitality of 95.9 and 92.7 per cent for the pansy and onion seed, respectively. The percentages of germination in the other cases, except the "B" sweet corn, pea, and bean, were sufficient to have produced a fair stand, i. e., if we consider that far too many seeds are usually sown. But a decrease in the percentage of germination means seeds of a low germinative energy. Even though the final percentage of germination be up to standard, the retardation may be of vital importance. A very good example of the retardation in germination is shown in the tests of the watermelon seeds. In the control sample 94 per cent of the seed germinated in 475 hours, while the seed returned from Mobile showed, during the same time, a germination of only 12 per cent; yet the difference in the final germination was only 0.3 per cent in favor of the control. Likewise the seed returned from San Juan germinated only 20 per cent in 47½ hours, the final germination being 96 per cent or only 2.3 per cent lower than the control.

Many similar cases might be mentioned in which the final percentages of germination, as shown by the first set of tests given in Table I, represent a loss such as might be justly considered well within the limits of normal variation. However, that all of the samples of seed were injured as a result of the unfavorable climatic conditions is shown in the second set of tests set forth in Table II. In the latter case the seeds remained in the various places nearly twice as long as those used for the first test.

Table II.—Effect of climate on vitality as shown by percentage of germination—second test.

Kind of seed.	Control.	Mobile, Ala., Feb. 17 to Nov. 6. 262 days.	Baton Rouge, La., Feb. 17 to Oct. 22. 247 days.	Dur- ham, N. H., Feb. 17 to Oct. 26. 251 days.	Au- burn, Ala., Feb. 17 to Nov. 19. 275 days.	Lake City, Fla., Feb. 9 to Oct. 1. 234 days.	Wag- oner, Ind. T., Feb. 17 to Oct. 13. 238 days.	San Juan, P. R., Feb. 9 to June 20. 129 days.	Ann Arbor, Mich.
Corn, sweet, "A"	94.5	20.0	88.0	96.0	88.0	92.0	90.0	92.0	98.0
Corn, sweet, "B"	88.5	12.0	54.2	82.0	62.0	77.0	78.0	78.0	80.0
Onion	97.0	0.0	0.5	0.0	12.0	16.5	24.5	50, 0	97.5
Cabbage	92.4	17.0	25, 5	12.0	61.5	63.5	70, 5	76.2	91.0
Radish	78.8	51.0	55, 5	59, 5	63.0	58.5	60.5	62.0	77.5
Carrot	82.0	8.5	25, 0	2.0	36.0	43.5	49.0	48, 5	86, 0
Pea	95, 7	44.0	80.0	91.0	97.9	86.5	80.0	98, 0	98.0
Bean	98.7	0.0	-60, 0	78.0	56.0	84.0	82.0	96, 0	100.0
Pansy	53.0	0.0	0.0	0.0	2.0	1.5	7.5	6, 5	46.5
Phlox drummondii	53.9	0,0	0.0	0.5	1.0	2.5	5.5	11.5	40.0
Tomato	97.5	79.5	96, 0	87.0	94.0	94.0	94.0	96.5	98.0
Watermelon	99.0	64.0	92.0	82.0	86.0	92.0	94.0	88.0	96.0
Lettuce	92.3	20.0	84,5	88.5	86.0	85.0	82.0	83.5	92.5
Average of all seeds.	86.77	24.31	50.86	52, 42	57.31	61.27	62.11	68, 21	81.58

Even though the columns in both Tables I and II are arranged in the order of the loss in vitality as shown by the averages of the various places, it will at once be seen that the relative degree of injury did not remain the same throughout the experiment. This is probably best explained by a variation in the climatic influences. It is evident that in some of the places where seeds were stored the effects were more deleterious during the time between the first and second tests than they were during the first period of storage of 128 days. The results given in Table II are of the greater value in showing the relative merits of the different localities as places for storing seeds, extending as they do over a longer period of time.

As a result of the second series of tests it was found that the average percentage of germination of all of the samples of seed that were stored in trade conditions at Mobile for 262 days was only 24.31 per cent. This is equivalent to a loss in vitality of 71.98 per cent as compared with the average percentage of germination of the control samples, the average germination of the controls being 86.77 per cent. The pansy, phlox, onion, and beans stored at Mobile wholly lost their power of germination. The tomato seed, which proved to be the most resistant to unfavorable conditions, gave a germination of 79.5 per cent, or a loss in vitality of 18.46 per cent, as compared with the control sample, which germinated 97.5 per cent. The degree of deterioration in the seeds stored at the other places was much less marked than for those stored at Mobile. The loss in vitality was only 41.39 per cent in the seeds returned from Baton Rouge. The results from the seeds which were stored at Durham, Auburn, Lake City, Wagoner, and San Juan differed but little from those from Baton Rouge. The relative losses in vitality are in the order given. The seeds kept in the packages which were stored under trade conditions in the laboratory at the University of Michigan showed a loss in vitality of only 2.52 per cent as compared with the control, the seeds of which were stored in a cool, dry closet on the fourth floor of the botanical laboratory. Ordinarily a loss of 2.52 per cent would be considered as a normal variation due to sampling and testing, and such was probably true in these two sets, with the exception of the greater deterioration of the phlox, pansy, and "B" sweet corn, which were undoubtedly injured by the unfavorable trade conditions, as repeated tests have shown.

From Table II it will also be seen that the "A" sweet corn, peas, tomato, and watermelon, with the exception of those returned from Mobile, show a fair percentage of germination. In some cases the final percentages of germination were even higher than the controls; but, as previously stated, the final germination is not always a good criterion for the determination of vitality, it being necessary to consider the germinative energy as a basis for comparison. In order to show this more fully some of the detailed results are herewith given in Table III. These results show to a good advantage the degree to which germination has been retarded.

Table III.—Retardation in germination due to injury caused by unfavorable climatic

	Corn	"A."	Pe	as.	Water	melon.	Tomato.			
Place where seeds were kept.	Germi- nation at end of 64 hours.	Final germi- nation.	Germination at end of 40 hours.	Final germi- nation.	Germination at end of 84 hours.	Final germi- nation.	Germination at end of 83 hours.	Germination at end of 107 hours.	Final germi- nation.	
	Per cent.	Per cent.	Per cent.	Per cent.	Per cent.	Per cent.	Per cent.	Per cent.	Per cen	
Control	81.3	91.5	79.6	95.7	98.0	99.0	78.0	92.7	97.	
Mobile, Ala	4.0	20.0	a 24. 0	41.0	0.0	64.0	1.5	12.5	79.	
San Juan, P. R	61.0	92.0	60.0	98.0	12.0	88.0	38.5	78.0	96.	
Baton Rouge, La	50.0	88.0	36.0	80.0	0.0	92.0	9.0	56.0	96.	
Wagoner, Ind. T	61.0	90.0	36.0	80.0	2.0	94.0	40.0	81.5	94.	
Lake City, Fla	68.0	92.0	50.0	86.0	0.0	92.0	16.5	65.0	94.	
Durham, N. H	86.0	96.0	54.0	91.0	0,0	82.0	0,5	5, 5	87.	
Auburn, Ala	80.0	88.0	a 93. 7	97. 9	22.0	86.0	59.0	75.5	94.	
Ann Arbor, Mich	82, 0	98.0	82.0	98. 0	91.0	96.0	75.5	91.0	98.	

a After 62 hours.

In order that the results of Tables I and II may be more readily and fully comprehended, it has been deemed advisable to summarize them in another table. For this purpose the average percentages of germination of all of the different samples of seed have been determined for each of the different places. From these average percentages of germination the deterioration in vitality, as shown by both the first and second tests as given in Tables I and II, have been calculated, the germination of the controls serving as a basis for comparison. These results furnish more trustworthy data as to the relative merits of the different localities as places for storing seeds. Likewise the percentages of deterioration between the time of the first and the second tests are shown in Table IV.

Table IV.—Average percentages of germination of all seeds kept at the various places, their deviations from the controls, and the increased percentages of loss in the second series of tests.

Place of storage.	tion of	germina- all seeds r experi-	Deteriors vitality pared v trols.	Deterio- ration in vitality between first and	
	First test.	Second test.	First test.	Second test.	second tests.
	Per cent.	Per cent.	Per cent.	Per cent.	Per cent.
Control	87.79	86.77			1, 16
Mobile, Ala	53, 59	24.31	38.95	71.98	54.61
San Juan, P. R	75.12	68, 21 a 45, 18	14 317	21.39 a 47.93	9, 20 a 39, 86
Baton Rouge, La	80.48	50.86	8.32	41.39	36.81
Durham, N. H.	85.57	52, 42	2,52	39.58	38.74
Auburn, Ala	85.70	57.34	2,38	33.91	33.10
Lake City, Fla	. 83.00	61.27	5.45	29, 38	26.18
Wagoner, Ind. T	82, 12	62.11	6, 45	28.41	21.37
Ann Arbor, Mich.	86, 23	81,58	1.77	2, 52	1.91

In Table IV the results are arranged in the order of the loss in vitality as shown by the second tests. However, a few words of explanation will be necessary, especially concerning the loss at San Juan. In the first place, the seeds were kept at San Juan only 131 days" during the early part of the summer, while during the most critical period, June 20 to November 6, they were in the botanical laboratory of the University of Michigan. Those marked Mobile, Ala., were, during the entire time, 262 days, under the influence of the warm, moist climate of the Gulf of Mexico. The seeds kept at other places can well be compared with those from Mobile, the time being approximately the same. The average loss as shown by the second tests was 3.35 times greater than the loss in the first test, which by calculation would bring San Juan next below Mobile, with a loss of vital energy in the seeds equal to 47.93 per cent. But more data are necessary before such a gradation of injurious climatic influences can be established.

Table IV, however, brings out another interesting point, as shown by comparing the results of the first and second tests at San Juan and Mobile. In the first test the loss in vitality of the seeds from Mobile was 38.95 per cent, while the seeds returned from San Juan showed a loss of only 14.31 per cent as compared with 71.98 and 21.39 per cent, respectively, as shown in Table II. The degree to which the seeds were injured while they were stored in San Juan was such that they continued to deteriorate much more rapidly than the control sample. This deterioration was most marked in the case of the pansy seed, the germination of the first test being 20 per cent and that of the second test only 6.5 per cent, showing a loss in vitality of 68.2 per cent and 87.7 per cent, respectively. Thus when seeds are once placed in conditions unfavorable for the preservation of their vitality for a sufficient length of time to cause some injury, this injury will always be manifest and cause a premature death of the seeds even though they afterwards be removed to more favorable conditions.

Seeds of strong vitality can withstand greater changes in conditions than seeds of low vitality without any marked deterioration. Throughout these experiments a wide difference has been observed between the "A" sweet corn and the "B" sweet corn. The original tests made January 30, 1900, at the time the seeds were received, showed a germination of 94 per cent for the "A" sample and 88 per cent for the "B" sample of corn. The control tests, made in November, 1900, showed a germination 0.5 per cent higher in each case; but the average loss in vitality of the two samples of seed kept at the various places was 12.17 per cent for the "A" sample and 26.10 per cent for the "B" sample. As with the pansy and the phlox these samples showed that

<sup>&</sup>lt;sup>a</sup>The number of days here given for San Juan is not absolutely correct. The time was reckoned from the date the seeds were sent from the laboratory until they were received in return.

the stronger the vitality of the original sample of seed the more harsh treatment can be undergone without being injured. Strong vitality implies long life as well as vigorous seedlings.

Another very important factor to be considered in the handling of seeds is the relative resistance of seeds of various species to adverse conditions. Certain seeds under one set of conditions may retain their vitality exceedingly well, while seeds of other species of plants under identical conditions may be killed in a comparatively short time. For this reason no general rule can be laid down for the preservation of seeds. Table V shows the varying degrees of deterioration of the different species of seeds used in the experiments.

Table V.—Different degrees of deterioration of various kinds of seeds.

		First test.			Second tes	t.
Kind of seed.	Germi- nation of control.	Average germination from the various places.	Deterio- ration in vitality as com- pared with the control samples.	Germi- nation of control.	Average germination from the various places.	Deterio- ration in vitality as com- pared with the control samples.
	Per cent.	Per cent.	Per cent.	Per cent.	Per cent.	Per cent.
Tomato	95, 5	93, 06	2.55	97.5	92, 43	5, 20
Pea	95.3	91,56	3.92	95.7	81.80	11.39
Corn, sweet, "A"	95. 9	94.75	1.20	94.5	83,00	12.17
Watermelon	98, 3	97.75	.57	99.0	86.62	12.51
Lettuce	81.6	80, 00	1.96	92.3	77.75	15,77
Radish	83.6	74.38	11.02	78.8	60.93	22, 67
Corn, sweet, "B"	89.3	78.16	12.47	88.5	65, 40	26.10
Bean	98.7	93.00	5, 76	98.7	69, 50	29.58
Cabbage	92.7	86,00	7, 22	92.4	52, 15	43.56
Carrot	83.3	75.16	9.77	82.0	37.81	53, 89
Onion	95, 8	82.18	15, 26	97.0	25.12	74.10
Pansy	63, 0	38, 87	38, 33	53.0	8,00	84.90
Phlox drummondii	69.0	44.87	34.97	53.9	7, 62	85, 85

In the above table the list of seeds is arranged in the order of their power to withstand the action of diverse climatic conditions, as shown by the results of the second test, given in Table II. Tomato seeds were found to be the most resistant, the control sample germinating 97.5 per cent. The average germination of the samples of tomato seed kept at the various places was 92.43 per cent, or a loss in vitality of only 5.20 per cent. The seed showing the next least injury was the peas, with a deterioration of 11.39 per cent. The phlox, which was the most affected by the unfavorable conditions, germinated only 7.62 per cent, thus showing a loss in vitality of 85.85 per cent.

It is also interesting to note that the order, as shown by the second series of tests, is quite different from that of the first. This lack of uniformity increases the difficulties that must be overcome before the causes of the loss of vitality in seeds can be fully comprehended. Were all seeds affected in the same way when subjected to identical con-

ditions, the order should have remained the same throughout, but the wide variation in atmospheric changes affects different seeds so very differently that no uniformity of results can be secured. For example, the conditions prevailing from February until June were much more disastrous to the vitality of the tomato and pea than to the "A" sweet corn, watermelon, and lettuce, while the conditions existing from June to November were more injurious to the "A" sweet corn, watermelon, and lettuce. An examination of the table will show other results of a similar nature. During the earlier stages of devitalization seeds undergo a gradual deterioration in vitality, but after reaching a certain stage in their decline there is a comparatively sudden falling off, and seeds, except perhaps a few of the most persistent, soon cease to show any power of germination. Such factors as these must be taken into account in determining the relative length of time that different kinds of seed will retain their vitality. But as yet sufficient information is lacking in order to make any trustworthy attempt to classify seeds in respect to their viable periods when subjected to different conditions. Numerous experiments are now under way, with the hope of furnishing a basis for such a classification.

In order to obtain more data as to the influence of climate upon vitality additional samples of seed were sent to Mobile and Baton Rouge, where they were stored under the same trade conditions as for the former experiment. For these tests only cabbage, lettuce, and onion seeds, put up in envelopes, as for the previous tests, were used. The different packages of seed, placed in paper boxes from which they were not removed, were sent from the laboratory on May 20, 1901, and were returned November 26, 1901, the total time of storage being 190 days. The results of these tests are shown in Table VI, and are even more striking than those of the former tests shown in Tables I and II.

Table VI.—Relative merits of Mobile, Ala., Baton Rouge, La., and Ann Arbor, Mich., as places for storing seeds.

	Cabbage.  Percentage of seeds germinated at the end of—			Lettuce.  Percentage of seeds germinated at the end of—			Onion.  Percentage of seeds germinated at the end of—			
Seeds subjected to "Trade condi- tions."										
	36 hours.	60 hours.	days.	36 hours.	60 hours.	11 days.	60 hours.	84 hours,	108 hours.	14 days.
Mobile, Ala	0.0	0.0	8.5	0.0	14.0	64.0	0.0	0.0	0.0	0.0
Baton Rouge, La	0.0	0.0	22, 5	2.5	35, 5	74.0	0.0	0.0	- 0.0	0.0
Ann Arbor, Mich	10.0	64.5	86.5	67.0	82.5	96.5	3.0	10.0	43.0	93.0

[Period, 190 days.]

Table VI shows quite clearly the deleterious action of the warm, moist climate of the Gulf of Mexico on the life of seeds. The onion seed which was stored at Mobile and Baton Rouge did not germinate,

while seed from the same lot stored at Ann Arbor germinated 93 per cent. The cabbage seed was injured nearly as much as the onion, the sample from Mobile germinating only 8.5 per cent. The conditions at Baton Rouge were slightly more favorable to the preservation of vitality. The cabbage seed stored at the latter place germinated 22.5 per cent, while a like sample of seed stored at Ann Arbor germinated 86.5 per cent. The lettuce was much more resistant than either the cabbage or the onion seed, but here, too, the injury was quite marked, especially as shown by the retardation in germination. The conditions at Mobile were also the most disastrous for the lettuce seed. During the first 36 hours that the tests were in the germinating chamber none of the lettuce seed from Mobile germinated, while the seed from the corresponding sample from Ann Arbor germinated 67 per cent. The final percentages of germination were 64 and 96.5 per cent, respectively, for the seed from Mobile and Ann Arbor, showing a loss in vitality of 33.68 per cent in the seed stored at Mobile. Here it will be seen, as in Table V, that the onion seed was most sensitive and the lettuce seed most resistant to the unfavorable conditions. In the first tests shown in Table V the average loss in vitality of the lettuce, cabbage, and onion was 15.77, 43.56, and 74.10 per cent, respectively, while for the last tests, as shown in the foregoing table, the losses in vitality of similar samples of seed kept at Mobile were 33.68, 91.29, and 100 per cent, respectively. The ratio is practically the same in both cases, the loss in the cabbage seed being 2.7 times greater than that of the lettuce.

The foregoing data are sufficient to indicate that climatic influences play a very important part in the life of seeds, and that the degree of injury varies greatly in different places and likewise in different seeds. Some seeds were practically worthless after an exposure of four or five months in such places as Mobile, Baton Rouge, or San Juan, as shown in Table I. After longer exposures, six or nine months, similar results were obtained from all of the places to which seeds were sent. Many of the seeds were killed, as shown in Table II. The conditions at Mobile were fatal to all of the seeds; that is, the seeds were worthless so far as the gardener is concerned.

#### CAUSES OF THE LOSSES IN VITALITY IN DIFFERENT CLIMATES.

Having shown that seeds lose their vitality much sooner in some localities than in others, the question naturally arises, "Why this loss in vitality?" Unfortunately only two of the places where seeds were stored, Mobile and San Juan, have Weather Bureau stations which are equipped for making complete observations of the meteorological conditions. It has been observed, however, that there is a very close relationship between the precipitation and the loss in vitality in seeds; that is to say, in a measure the loss in vitality is directly proportional to the amount of rainfall. This deterioration is more apparent as the

temperature increases, but the injury due to the increase in temperature is dependent on the amount of moisture present.

The following table has been compiled in order to show the ratio between the loss in vitality and the precipitation and temperature. The loss in vitality, as given in the second column of Table VII, represents the average losses in percentages, calculated from the results of the germination tests of the 13 different samples of seeds, as shown in Table II. <sup>a</sup>

The third column shows the annual precipitation in inches. The annual precipitation has been taken, because in some instances heavy rainfalls occurred just previous to the time that the seeds were put into storage. Then, too, the annual precipitation furnishes more accurate data for a basis of comparison. The mean temperatures, as given in column 4, are not the mean annual temperatures, but the averages covering the time during which the seeds were stored. The mean annual temperatures were not taken, chiefly for the reason that the critical period, in so far as temperature is concerned, is during the summer months.

Table VII.—Ratio between vitality, precipitation, and temperature. b

Place where seeds were stored,	Average loss in vi- tality of the 13 dif-	Annual precipita-	Tempe	rature.
	ferent sam- ples of seeds.	tion.	Mean Fahr.	Maximum Fahr.
	Per cent.	Inches.	Degrees.	Degrees.
Mobile, Ala	71.98	91.18	71.4	96.0
Baton Rouge, La	41, 39	66.37	72.2	98.0
Durham, N. H	39, 58	48.20	52.3	98. 0
Auburn, Ala	33, 91	62, 61	64.4	98.0
Lake City, Fla	29, 38	49.76	73.3	103.0
Wagoner, Ind. T	28, 41	42.40	67.1	167.0
Ann Arbor, Mich	2.52	28.58	49.12	98.0

a These seeds were sent out in February, 1900, and were returned to the botanical laboratory and tested in October and November, 1900. The average time that the seeds were kept at the various places was 252 days.

b The results of the San Juan tests have been omitted from this table because, as has been previously stated, all of the seeds were returned from San Juan on June 20, 1900, when the first tests were made. The second series of tests was made in October, 1900. During the time intervening between the first and second tests the San Juan samples were kept in the botanical laboratory at the University of Michigan.

According to the table the seeds kept at Mobile suffered the greatest loss in vitality. However, it is quite probable that the greatest loss would have been from the seeds stored at San Juan had the time of storage been the same for the two places, so that the results of the San Juan tests could have been included in the table. This conclusion is based on the following facts: Normally, the number of rainy days at San Juan far exceeds those at Mobile. In 1900 there were 211 days on which rain fell in San Juan, while the records for Mobile show only 146. Likewise the average temperature of the dew-point for San Juan was 71° F. and only 59° F. for Mobile, which, when expressed in terms of absolute moisture, gives 8.240 and 5.555 grains of water per cubic foot at the time of saturation. On the other hand, the relative humidity of San Juan was 78.5 per cent, or slightly lower than that of Mobile, the latter having a relative humidity of 80.5 per cent. However, the mean annual temperatures were 77.6° and 71.4° F., respectively, hence a mean absolute humidity of 7.099 grains of aqueous vapor for San Juan and only 6.718 grains per cubic foot for Mobile.

From the foregoing table it will be seen that precipitation is a factor of much greater importance than temperature. In order to show the real value which the amount of precipitation furnishes as a basis for judging the length of time that seeds will retain their vitality when stored in localities having a marked difference in the amount of rainfall, the results set forth in the above table are represented diagrammatically as follows:

Effect of precipitation on vitality.

Place.	Percentage of loss in vitality.	Inches of precipitation.
Mobile	71.98	91.18
Baton Rouge	41.39	66.37
Durham	39.58	48.20
Auburn	33.91	62.61
Lake City	29.38	49.76
Wagoner	28, 41	42.40
Ann Arbor	2.52	28, 58

A discrepancy is very marked for Durham, N. H., which may be partially explained by considering again the conditions under which the seeds were stored. It will be remembered that these samples of seeds were stored in a hall which opened directly into a chemical laboratory. It is quite probable that the low percentages of germination were due to the injurious action of gases emanating from the laboratory. Of these gases, ammonia probably played a very important part, as it is well known that seeds are very readily injured when subjected to the action of ammonia.

It is to be understood that the above comparisons are somewhat indefinite. If the amount of rainfall were equally distributed throughout the year a definite ratio could, in all probability, be established; but in the majority of places there are alternating wet and dry seasons, which make such a comparison very difficult and unsatisfactory. Yet for ordinary considerations it is sufficient to say that seeds will retain their vitality much better in places having a small amount of rainfall. For more exact comparison other factors must be taken into account, especially the relative humidity, mean temperature, and temperature of the dew-point, which ultimately resolves itself into the absolute amount of moisture present in the atmosphere.

#### EFFECT OF MOISTURE AND TEMPERATURE UPON VITALITY.

From the foregoing experiments it is quite evident that moisture plays an important part in bringing about the premature death of seeds and that the detrimental action of moisture is more marked as the temperature increases. Formerly the general consensus of opinion has been to make this statement in the reverse order—that is, that temperature exerts a very harmful action on seeds if much moisture be present. For comparatively high temperatures the latter statement would probably suffice—at least it is not misleading, and in a certain measure it is true; but at the lowest known temperatures, as well as at ordinary temperatures, moisture is the controlling factor, and in order to be consistent it should likewise be so considered for higher temperatures—that is, within reasonable limits.

That temperature is only of secondary importance is brought out in the results obtained by a number of investigators. It has been well established by Sachs, Haberlandt, Just, Krasau, Isidore-Pierre, Jodin, Dixon, and others that most seeds, if dry, are capable of germination after being subjected to relatively high temperatures for periods of short duration. The maximum for most seeds is a temperature of 100° C. for one hour; but if the seeds contain comparatively large quantities of moisture they are killed at much lower temperatures. It has been reported that lettuce seed will lose its vitality in two weeks in some of the tropical climates where moisture is abundant. Dixon has shown that if lettuce seed be dry it will not all be killed until the temperature has been raised to 114° C.

In case of low temperatures the factor of moisture is of less importance, yet even under such conditions the moisture must not be excessive or the injury will be quite apparent. But if seeds are well dried it can safely be said that they will not be killed as a result of short exposures to the lowest temperatures which have thus far been produced. Our knowledge of the resistance of seeds to extremely low temperatures is based on the experiments of Edwards and Colin, Wartmann, C. De Candolle and Pictet, Dewar and McKendrick, Pictet, C. De Candolle, Brown and Escombe, Selby, and Thiselton-

a Handbuch d. Exp. Phys. d. Pflanzen, Leipzig, 1865, p. 66.

b Pflanzenbau I, 1875, pp. 109–117; Abs. in Bot. Jahresbr., 1875, p. 777.

<sup>&</sup>lt;sup>c</sup> Bot. Zeit., 33, Jahrg. 1875, p. 52; Cohn's Beiträge zur Biol. der Pflanzen, 1877, 2: 311–348.

<sup>&</sup>lt;sup>d</sup>Sitzungsbr. d. Wiener Akad. d. Wiss., 1873, **48**: 195–208. I. Abth.

<sup>&</sup>lt;sup>c</sup>Ann. Agron., 1876, 2: 177–181; Abs. in Bot. Jahresbr., 1876, II. Abth., 4: 880.

f Compt. Rend., 1899, 129: 893-894.

<sup>#</sup> Nature, 1901, **64**: 256–257; notes from the Botanical School of Trinity College, Dublin, August, 1902, pp. 176–186.

<sup>&</sup>lt;sup>h</sup> Ann. sci. nat. bot., ser. 2, 1834, 1: 257–270.

i Arch. d. sci. phys. et nat., Genève, 1860, 8: 277-279; ibid., ser. 3, 1881, 5: 340-344.

J Ibid., ser. 3, 1879, 2: 629-632; ibid., ser. 3, 1884, 11: 325-327.

k Proc. Roy. Inst., 1892, 12: 699.

l Arch. d. sci. phys. et nat., Genève, ser. 4, 1893, 30: 293-314.

m Ibid., ser. 4, 1895, 33: 497-512.

<sup>&</sup>lt;sup>n</sup> Proc. Roy. Soc., 1897-8, **62**: 160-165.

o Bul. Torr. Bot. Club., 1901, 28: 675-679.

Dyer." In the experiments of the last-named investigator seeds were subjected to the temperature of liquid hydrogen (-250 to -252 C.) for six hours, and when tested for vitality the germination was perfect and complete. <sup>b</sup>

Much more might be said on the effect of high and low temperatures on vitality. But for the commercial handling of seeds the extremes of temperature are of secondary importance and need not be further discussed at this time. In the present work the purpose has been to show the effect of moisture on the vitality of seeds when subjected to such temperatures as are usually met with in the storing of seeds.

#### SEEDS PACKED IN ICE.

On February 6, 1900, samples of each of thirteen kinds of seed were put up in duplicate, both in manila coin envelopes and in small bottles. The bottles were closed with carefully selected cork stoppers. These two sets of duplicate samples were then divided into two lots. Each lot contained one of each of the packages and one of each of the bottles of seeds. The samples thus prepared were carefully packed with excelsior in wooden boxes, the boxes being then wrapped with heavy manila paper. In one of the boxes was also placed a Sixes' self-registering thermometer, so that the minimum temperature could be ascertained.

These boxes were stored in a large ice house near Ann Arbor, being securely packed in with the ice at the time the house was being filled. The first box was taken out with the ice on June 12, 1900, after a lapse of 126 days. The thermometer in this box registered a minimum of -3.6°C. It is safe to assume that this temperature was uniform, at least up to within a few days of the time when the seeds were taken out. Unfortunately, absence from the university at this particular time delayed an examination of the seeds until June 20. During the eight intervening days the box of seeds was kept in the laboratory and there many of the seeds in the packages molded, so that they were unfit for germination tests. In fact, the results of the tests from the packages are of little value within themselves; but in comparison with the vitality tests of the seeds kept in the bottles some important facts are brought out, and it has been deemed advisable to tabulate these results with those of the second series.

The second box of seeds was packed approximately in the center of a large ice house (100 by 60 by 20 feet) and was taken out with the ice on July 21, 1900, after having been 167 days in cold storage. The

a Proc. Roy. Soc., 1899, 65: 361-368.

b Brassica alba (oily), Pisum sativum (nitrogenous), Cucurbita pepo (oily), Triticum sativum (farinaceous), and Hordeum vulgare (farinaceous).

box was brought directly to the laboratory and the seeds were examined at once. Those contained in the paper packages had absorbed a considerable quantity of moisture and were much softened. In all of the packages except those containing the onion and watermelon seeds some mold had developed; but in the seeds used for the germination tests care was taken to avoid using those that showed any trace of a mycelium, thereby reducing the injury due to fungous growth to a minimum, even though subsequent experiments have shown that such injury is practically negligible.

An interesting point concerning the germination of some of the seeds at this low temperature may be stated in this connection. Eight of the peas, or 4 per cent, had already germinated, the radicles varying in length from 1 to 2.5 cm., thus corroborating Uloth's results in germinating peas at or slightly below the temperature of melting ice.

Table VIII.—The vitality of seeds kept in an ice house in envelopes and bottles, and likewise the vitality of the controls.

	1	First test	, after 1	26 days.		Second test, after 167 days.					
	Germination.			Differ-	Differ- encebe-	Ge	rminati	Differ- ence be- ence be-			
Kind of seed.	Con- trol,	Envel- ope.	Bottle.	tween envel- ope and control sam- ples.	tween envel-		Envel- ope.	Bottle.	tween envel-	tween envel- ope and	
	Per ct.	Per ct.	Per ct.	Per ct.	Per ct.	Per ct.	Per ct.	Per et.	Per ct.	Per ct.	
Corn "A"	96.0	36.0	94.0	60.0	58, 0	92.0	86.0	96.0	6.0	10.0	
Corn "B"	90,0	60.0	96.0	30.0	36.0	92.0	74.0	94.0	18.0	20.0	
Onion	95.0	92.5	96, 5	2.5	4.0	95.0	94.5	95.0	0.5	0,5	
Cabbage	93, 5	89.0	94, 0	4.5	5.0	92.0	90.0	94.0	2.0	4.0	
Radish	88, 5		81.5			80.5	74.0	89.0	6.5	15.0	
Carrot	79, 5		80.0			73.5	52.0	75.5	21.5	23.5	
Pea	92.0		88.0			94.7	90.0	96.0	4.7	6,0	
Bean	100.0		100.0			100.0	0.0	98.0	100.0	98.0	
Pansy	52.5	5.0	65, 5	47.5	60.5	52.0	2.5	65, 5	49.5	63.0	
Phlox	74.0		a 16.5			54.0	11.0	68.5	43.0	57.5	
Tomato	95.5	73.0	93.5	22.5	20, 5	96.5	51.5	96.0	45, 0	44.5	
Watermelon	\$8.0	90.0	100.0	8.0	10.0	100,0	96, 0	100.0	4.0	4.0	
Lettuce	80.0		66.0			81.5	66, 0	71.0	15.5	5.0	
Average	87, 3	63.6	87.9	25.0	27.7	84.9	62.1	87.6	24.3	27.0	

aIn making up the averages the result of the germination of the phlox was omitted because a subsequent examination showed that the bottle containing this sample of seed was broken at the bottom, thus admitting sufficient moisture to destroy vitality, as is borne out by the second test.

The above table shows, as previously stated, that the results of the first tests are incomplete and not very satisfactory, owing to the fact that the germination tests were unavoidably delayed for eight days after the seeds were taken from the ice house; but with the second set

of samples the counts for the vitality tests were begun within an hour from the time the seeds were removed from the ice house. Thus, the conclusions for these experiments must be drawn chiefly from the second series of tests. However, comparisons will be made with the first where such seem justifiable.

It will at once be seen that the seeds which were in paper packages gave a much lower percentage of germination than either the control samples or those kept in bottles. The average germination of the controls was 84.9 per cent, and the average germination of the seeds kept in bottles was 87.6 per cent, while only 62.1 per cent of the seeds kept in paper packages germinated. This is equivalent to a loss in vitality of 24.3 and 27 per cent, respectively, as compared with the vitality of the control samples and the samples from the bottles. The results of the first tests are practically the same, save that the differences between the control and the bottle samples are less marked. In the second case the average vitality of the seeds kept in envelopes was much reduced by the complete failure to germinate in the case of the beans, which are most susceptible to the deleterious action of moisture at the given low temperature.

One of the most important points brought out by these experiments is the result obtained with onion, cabbage, and watermelon seeds. both the first and the second tests the germination varied but little throughout. However, in all cases the seeds in the paper packages were slightly injured by the action of the moisture. This factor is of much importance, especially in the case of the onion seed, which, when kept in a moist atmosphere at normal temperatures, soon loses its vitality, but when maintained at temperatures slightly below freezing it becomes very resistant to the action of moisture. The beans, on the other hand, were all killed, although they are ordinarily much more hardy than onion seed. It is quite probable, however, that the death of the beans may be attributed to the reduction in temperature. Containing as they do large quantities of starch, they absorb more water than less starchy or more oily seeds. This factor, together with the large embryo, renders them much more susceptible to the injurious action of freezing temperatures.

Another important feature brought out by these experiments was the better germination of the seeds which had been stored in bottles in the ice house. The average germination of these samples was 2.7 per cent higher than that of the control. In a measure this may be included within the limits of variation; but when it is considered that all of the bottle samples except the beans, tomato, and lettuce showed a vitality equal to or greater than the control, it can hardly be considered as a normal variation, especially since only the lettuce gave any marked variation in favor of the control. Likewise, the average percentages

of the first series of tests show a slight increase in favor of the seeds kept in the bottles, though the increase is not so well marked and is less uniform than in those of the second series.

Aside from the final germination there is still another factor that must be taken into consideration as bearing evidence of the advantage of keeping seeds at low temperatures, provided that they are kept dry. All of the samples that were stored in the ice house in bottles showed a marked acceleration in germination. It is quite evident that the respiratory activities and accompanying chemical transformations were much reduced by the reduction in temperature, and the vital energy was thus conserved; but when the conditions were favorable for germination the greater amount of reserve energy in these seeds gave rise to a more vigorous activity within the cells and a corresponding acceleration in germination.

Numerous other experiments showing the effect of moisture on the vitality of seeds were made. In contrast to those just given, the injurious action of moisture at higher temperatures, yet temperatures well within the limits of those ordinarily met with in the handling of seeds, will be next considered.

#### EFFECT OF MOISTURE ON VITALITY AT HIGHER TEMPERATURES.

This set of experiments was undertaken particularly to furnish conditions somewhat similar to those existing in the States bordering on the Gulf of Mexico, or, in fact, all places having a relatively high degree of humidity and a temperature ranging from 30 to 37 °C. (86° to 98.6° F.) during the summer months. In order to secure the desired degrees of temperature two incubators were utilized, one being maintained at a temperature varying from 30 to 32 °C., the other from 36° to 37° C. The thermo-regulators were so adjusted as to admit of a possible variation of nearly two degrees in each case.

Beans, cabbage, carrot, lettuce, and onion were used for these tests. In each of the incubators the seeds were subjected to four different methods of treatment: 1. In a moist atmosphere, in free communication with the outside air. 2. In a moist atmosphere, but not in contact with fresh air, the seeds being in sealed bottles of 250 cc. capacity. 3. In a dry atmosphere, in free communication with the outside air. 4. Air-dried seeds in sealed bottles.

In order to obtain the conditions requisite for the first method of treatment, an apparatus was used as shown in figure 1. The seeds were put up in small packages and then placed in a 250 cc. bottle. The bottle containing the packages of seeds was placed within a specimen jar which was partially filled with water. This jar was then closed with a large cork stopper which carried two glass tubes, each of 1 cm. bore. These tubes extended 25 cm. above the top of the jar and out through

the opening in the top of the incubator. The primary object of the tubes was to prevent any water vapor from escaping within the incubator and thereby doing damage to the seeds that were to be kept dry

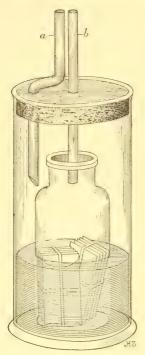


Fig. 1.—Apparatus used to determine the effect of moisture and temperature on the vitality of seeds in communication with free air.

in the same incubator. For the same reason the cork in the jar was well coated with paraffin. Approximately the same volume of water was maintained in the jar throughout the experiment, more water being added through tube a, as occasion demanded, to replace the loss by evaporation. The chief advantage in having two tubes was the comparative ease with which the air within could be displaced by a fresh supply by forcing a current of fresh air through one or the other of the tubes.

Two such preparations were made, one being left in the oven maintained at a temperature

varying from 30° to 32° C., the other in the oven maintained at a tempera ture varying from 36° to 37° C. In both cases the bottles contained five packages of each of the five samples of seed, thus making provisions for testing at different intervals.

In order to supply the conditions for the second method of treatment,

similar packages from the same samples of seeds were put into 8-ounce bottles, which were then kept for five days in a moist chamber. The increase in weight due to the absorption of water within the five days was as follows: Beans, 3.03 per cent; cabbage, 8.09 per cent; carrot, 8.26 per cent; lettuce, 7.45 per cent, and onion 8.43 per cent. This increase, with the water already present in the air-dried seeds, gave a water content of 13.23 per cent for the beans, 13.99 per cent for the cabbage, 13.60 per cent for the carrot, 12.45 per cent for the lettuce, and 14.84 per cent for the onion.



FIG. 2.—Apparatus used to determine the effect of moisture and temperature on the vitality of seeds not in communication with free air.

The bottles were then corked and sealed with paraffin, but were so

constructed that the relative humidity of the inclosed air could be increased without the admission of more free air. The detailed construction of this apparatus is shown in fig. 2.<sup>a</sup>

The seeds continued to absorb moisture to a limited extent. In order that the inclosed air might be maintained at approximately the same degree of saturation, a crude hygroscope was attached on the inside of each bottle. These hygroscopes were made from awns of Stipac capillata L., the tip of the awns being removed and a short piece of fine copper wire used as an indicator. These hygroscopes were suspended from the under side of the cork, as shown at h, and by the side of each was suspended a fine fiber of silk, which, being carried around by the indicator, recorded the number of turns made by the awn.

Five such preparations were made for each of the two sets, so as to furnish seeds for a series of tests. One set was kept at a temperature of 30° to 32° C, the other at 36° to 37° C. The seed from one of the bottles, at each of the temperatures, was weighed after eighty-one days, at the time the germination tests were made. These weighings showed that at the lower temperatures the average increase in weight for all the seeds was 8.6 per cent, and at the higher temperatures, 6.3 per cent. The increase in the case of the beans was quite marked at this time, being 13.3 per cent for those maintained at a temperature ranging from 30° to 32° C, and 9.8 per cent for those maintained at 36° to 37° C.

The third set of conditions consisted simply of packages of the airdried seeds kept in open boxes in each of the incubators. This series of tests was made especially for the purpose of determining the effect of dry heat on the vitality of seeds when maintained at the temperatures above given for some considerable time.

For the fourth series small packages of the seeds were put into 2-ounce bottles, which were then corked and sealed with paraffin. Five of these bottles were kept in each of the ovens and germination tests were made at irregular intervals. The results of these tests furnish a

a The wide-mouth bottle (b) contains the packages of seed (s). Through an opening in the cork is inserted a short piece of soft glass tubing, being first fused at the lower end and having a slight constriction drawn at c. At a distance of 1 cm. above the constriction is blown a small opening, as shown at o. A short piece of heavy rubber tubing (t), cemented on a piece of heavy brass wire (w), serves as a stopper. This stopper, which must fit closely within the glass tube, is operated by means of the heavy wire. When drawn up, the water in the tube may give off aqueous vapor, which can escape through the small opening (o) into the bottle. When sufficient moisture is present the supply is shut off by pushing the stopper down firmly against the constriction. The stopper must be well coated with vaseline to prevent its sticking to the sides of the glass tube. To make the apparatus more secure against the entrance of fresh air, a second piece of rubber tubing (r) is placed in the upper part of the glass tube, the top of which is then filled with oil.

basis for comparing the relative merits of keeping seeds in open vessels and in sealed bottles.

Table IX will show the effect of the various methods of treatment on the vitality of the seeds.

Table IX.—Vitality of seeds when subjected to the action of a dry and a moist atmosphere, both when exposed to free air and when confined in glass bottles, at relatively high temperatures. <sup>a</sup>

	Begin- ning of experi- ment.	End of experi- ment and date of germina- tion tests.		Dura- tion of ex- peri- ment.	Vitality of seeds when kept in a moist at- mosphere.				Vitality of seeds when kept in a dry atmosphere.				Ger-
Kind of seed.					tles, at tem- peratures				In open boxes, at tempera- tures vary- ing from—		In sealed bottles, at tempera- tures vary- ing from—		tion of con- trol sam- ples,
					30° to 32°.	36° to 37°.	30° to 32°.	36° to 37°.	30° to 32°.		30° to 32°,		1,1000
				Days.	P. ct.	P. ct.	P. ct.	P. ct.	P. ct.	P. ct.	P. ct.	P. ct.	P. ct.
Bean	Mar. 4	Apr.	4	31	100.0	100.0	78.0	44.0	86.0	84.0	98.0	98.0	94.0
Do	do	May	12	69	97.5	0.0	75.0	0.0	100.0	90.0	92.5	95.0	98.7
Do	do	May	24	81	94.0		0.0	0.0	98.0	90.0	98.0	100.0	98.0
Do	do	July	22	140	2.3		0.0	0.0	100.0	94.0	98.0	96.0	99.4
Cabbage	do	Apr.	4	31	87.8	90.5	73.0	72.5	86.5	81.0	83.5	86.9	91.0
Do	do	May	12	69	71.6	0.0	30.0	0.0	67.5	87.9	79.0	78.5	83.0
Do	do	Мау	24	81	80.0		1.0	0.0	89.0	92.0	92.5	92.0	92.5
Do	do	July	22	140	0.0		0.0	0.0	84.0	83.0	88, 5	86.7	93.1
Carrot	do	Apr.	4	31	83, 5	77.5	54.5	29.5	84.5	88.0	89, 5	89.0	92.5
Do	do	May	12	69	69, 5	0.0	22.5	0.5	82.0	85, 0	83.5	82.5	78.0
Do	do	May	24	81	48.0		2.5	0.0	44.5	50.0	50.0	48.0	61.5
Do	do	July	22	140	0.5		0.5	0.0	81.0	81.2	78.5	83.1	83.1
Lettuce	do	Apr.	4	31	92, 5	90.5	78.0	58.0	91.0	86.5	91.5	90.0	90.0
Do		1	12	69	38.0	0.0	44.5	2.0	42.0	38.5	38.5	51, 5	31,5
Do		May		81	55, 5		1.0	0.0	65, 0	58.5	62, 5	67.0	53.5
Do	do	July		140	0.0		1.5	0.0	82.0	87.0	81.5	88.0	79.9
Onion	o.do	Apr.	4	31	95.5	89.0	64.5	45.0	95, 5	. 93, 0	96, 0	97.5	96, 0
Do		May		69	68.0	0.0	2.5	0.0	97.0	95, 0	97.5	93. 0	98, 5
Do		May		81	59.5		0.0	0.0	95.5	94.0	99.0	95.0	96.5
Do		-		140	0.0		0.0	0.0	90.0	92.0	97.5	94.7	95. 4
		1		1 10				0.0	00.0				

aA study of the table will show that the lettuce and carrot seed germinated very poorly at the end of 69 and 81 days. This, however, was not due to any inherent quality of the seed, but to an excessive temperature at the time the tests were made. Both of these seeds require a comparatively low temperature for their successful germination, lettuce germinating best at 20° C., and carrot at an alternating temperature of from 20° to 30° C.

The amount of moisture absorbed or expelled under the different methods of treatment has an important bearing on the duration of vitality and will be considered briefly at this time. Only the general results will be discussed in this connection, inasmuch as later experiments, carried out in a similar manner, show the detailed results to much better advantage. Nevertheless, it requires only a glance at the above table to show the marked difference in the germinative power of seeds which have been stored in moist and in dry conditions. The seeds which were exposed in a moist atmosphere to the higher

temperatures (36 to 37 C.) were killed much earlier than those subjected to the moist atmosphere at the lower temperatures 30 to 32° C.—in both the open and the closed bottles.

A weighing at the end of 31 days showed that the average increase in weight of the seeds kept in the open, moist chamber, due to the absorption of moisture, was 6 per cent at a temperature of 30 to 32° C., and 5 per cent at a temperature of 36° to 37° C. For the seeds kept in the oven, maintained at the temperature of 30° to 32° C., another weighing was made at the end of 134 days, at which time the average increase in the water content had risen to 8.67 per cent. Unfortunately the seeds from the second oven, maintained at the higher temperature, had become badly molded in 69 days, so that only the one weighing was made.

Vitality tests made at this time, 69 days, showed that all of the seeds from the open, moist chamber, at the higher temperatures, had been previously killed as a result of the drastic treatment; consequently no future germination tests were made. Those maintained at the lower temperatures were almost entirely free from mold at the expiration of the experiment, only an occasional seed showing any trace of fungous growth. Nevertheless, germination tests showed that the vitality had been destroyed in the cabbage, lettuce, and onion. Beans and carrot were most resistant, the former having germinated 2.3 per cent and the latter 0.5 per cent. All of the seeds had become very much softened. The beans and the lettuce had changed very materially in color, the beans (Early Kidney Wax Six Weeks) having become much darker and the lettuce (Black-Seeded Simpson) almost a lemon color.

With the seeds constituting the second series, i. e., in a moist atmosphere but in scaled bottles, the injury was much more severe. Here, as with the open chambers, the seeds subjected to the higher temperatures were killed first, even though the amount of moisture actually absorbed was less, as was also true with the other series. A weighing made at the end of 81 days gave an increase of 8.6 per cent for those from the oven maintained at a temperature of 30° to 32°C., and 6.3 per cent at the higher temperature. Likewise, in this series, the seeds had become very much softened and a very disagreeable odor had developed as a result of the putrefaction of their nitrogenous constituents. A close examination made at the end of 81 days revealed slight traces of fungous growth, but there is no reason to believe that these played any part in the destruction of vitality. However, in making counts for germination tests all molded seeds were carefully discarded.

The results of the germination tests showed that the vitality of the seeds kept at the lower temperatures had been practically destroyed at this time. The beans and onions failed to germinate, while the

cabbage, carrot, and lettuce germinated only 1, 2.5, and 1 per cent, respectively.

During the succeeding 60 days much mold had developed, and at the expiration of the experiment, 140 days, only the carrot and the lettuce gave any indications of vitality. It is especially interesting to note with what rapidity the deterioration took place between the sixty-ninth and the eighty-first days, showing that when vitality reaches a certain point in its decline there follows a comparatively sudden death. This same fact is also shown in the case of those seeds in this same series kept at the higher temperature. After 31 days' treatment they all failed to germinate, except 0.5 per cent in carrot and 2 per cent in lettuce seeds.

In the two series of experiments just considered there was an increase in water content as a result of the humidity of the air in which the seeds were kept. But the third series, open and dry, presents quite another factor. A weighing made at the end of 30 days showed that there had been an average loss of 2.5 per cent for the lower temperatures and 3.5 per cent for higher temperatures. After this time the weight remained nearly constant. Subsequent experiments, which will be considered later, also show that the water capable of being expelled at any given atmospheric temperature is driven off in a comparatively short time. In case of seeds this condition is practically completed in eight or ten days when maintained at temperatures as above given. This extra drying of the seed causes a greater contraction of the seed coats, and in a number of cases a corresponding retardation in the rapidity with which germination takes place. The retardation in the germinative activity is dependent on the increased difficulty with which the seeds absorb water, and in many cases has an important bearing on the vitality tests.

The fourth and last series, in which the air-dried seeds were sealed in bottles and subjected to the temperatures at which the two ovens were maintained, gave still another very different set of conditions. Here there was also an increase in weight, due probably to some process of oxidation, but the increase was very slight. The average increase from those kept at either of the temperatures was less than one-half of one per cent.

Seeds, if well matured and thoroughly air-dried, are not injured when kept at temperatures below 37° C., whether they be kept in free communication with fresh air, or in sealed bottles, or tubes. In the experiments under discussion the average percentage of germination was slightly higher in the ease of the seeds which had been stored in the sealed bottles. The mean percentage of germination for the seeds which had been exposed to the open air at a temperature of 30° to 32° C. was 83.05 per cent. Those from the sealed bottles kept at the same temperature germinated 84.82 per cent. At the higher temperatures—36° to 37° C.—the mean germination of the seeds from the open

and the closed bottles was 82.68 and 85.62 per cent, respectively. The control sample germinated 85.45 per cent. That 37° C, is about the maximum temperature at which air-dried seeds can be stored without injury is shown by the following experiments.

Preparations similar to those above mentioned were used, and after being subjected to a temperature of 37° C. for 219 days, there was no appreciable loss in vitality, except the deterioration of 4 per cent in the case of the cabbage seed that was kept in an open bottle, and 6.3 per cent in the seed from a closed bottle." But by increasing the temperature, during an additional period of 68 days, from 37° C. to a maximum of 44° C., the injury was much more marked, especially in the closed bottles. In the open bottles the vitality of the cabbage was lowered from 91.3 per cent to 77 per cent, representing a loss in vitality of 15.66 per cent. The onion seed fell from 95.7 per cent to 87 per cent when kept in an open bottle, and to 61 per cent when kept in a closed bottle. The beans showed no apparent injury in either case, except that they became very dry; consequently there was a retardation in germination as a result of the slow absorption of water.

The greater loss in vitality of the seeds kept in the bottles was the direct result of the higher humidity of the air immediately surrounding the seed, and not because there was a deficiency in the supply of fresh air, as might be readily assumed. In the open receptacles the additional amount of free water expelled, as a result of the increase in temperature, was allowed to escape, while in the sealed bottles it only gave rise to a relatively moist atmosphere, and consequently to a premature death of some of the seeds. If seeds are to be so confined, they should be previously dried at a temperature at which they are to be stored.

All of these seeds had become very dry and brittle. The odor of the air confined within the sealed bottles had become very unpleasant; likewise there was a marked change in the color of the seed coats of the inclosed seeds.

#### SUMMARY.

Most seeds if kept dry are not injured by prolonged exposures to temperatures below 37° C. (98.6° F.), it being immaterial whether they are in open or in sealed bottles.

If the temperature be increased above 37° C., vitality is seriously reduced.

If seeds are kept in a moist atmosphere, a temperature even as high as 30° C. (86° F.) works much injury in a comparatively short period. The degree of injury rapidly increases as the temperature rises.

Provided the degree of saturation is the same, the deleterious effect of moisture is fully as great in open as in closed bottles.

<sup>&</sup>quot;Only cabbage, onion, and beans were used for this experiment, the carrot and the lettuce seed being omitted.

# THE EFFECT OF DEFINITE QUANTITIES OF MOISTURE ON THE VITALITY OF SEEDS WHEN THEY ARE KEPT WITHIN CERTAIN KNOWN LIMITS OF TEMPERATURE.

The results of the experiments just discussed furnish a fair criterion by which to judge the vitality of seeds when influenced by temperature and moisture. It was still necessary to determine the effect of definite quantities of moisture on the vitality of seeds when they are submitted to temperatures well within the limits of that which may be encountered in commercial transactions.

On December 19, 1900, preparations were made to determine these factors. Seeds of cabbage, lettuce, onion, tomato, and peas were used for these experiments, which continued for 70 or 72 days. All of this seed was of the harvest of 1899 and had been in the laboratory during the eleven months immediately preceding the setting up of the experiments, being thus thoroughly air-dried. The amount of moisture present in the seeds at this time, as indicated by drying at 100° C., was as follows: Cabbage, 5.90 per cent; lettuce, 5 per cent; onion, 6.41 per cent; tomato, 4.71 per cent, and peas, 8.44 per cent.

The preparations were made as follows:

(a) Air-dried seeds were placed in bottles of 125 cc. capacity. The bottles were closed with cotton plugs in order to protect the seeds from dust while permitting a free circulation of air. This set served largely as a check.

(b) Air-dried seeds were carefully weighed and then put into 125 cc.

bottles, closed with firm corks, and sealed with paraffin.

 $(c, d, \epsilon, \text{ and } f)$  These samples were also carefully weighed and scaled in bottles as b, but in the different series of bottles there was first introduced 0.5, 1, 2, and 3 cc. of water which had been previously absorbed by small strips of filter paper.

(g) The seeds constituting this series were first dried for 30 days at a temperature of from 30 to 32 °C, and then put up in bottles which were sealed with paraffin. The loss in weight as a result of the drying was as follows: Cabbage, 2.41 per cent; lettuce, 2.59 per cent; tomato, 2.71 per cent, and onion, 3.47 per cent, leaving a water content of only 3.49 per cent, 2.41 per cent, 2 per cent, and 2.94 per cent, respectively. (Peas were not included in this series.)

One of each of the above preparations was then subjected to different

degrees of temperature as follows:

(1) Outdoor conditions, protected from rain and snow, but freely subject to all changes in temperature and humidity. The temperature during the time of the experiment, December 19, 1900, to February 28, 1901, varied from a minimum of  $-21.6^{\circ}$  C. to a maximum of  $8.9^{\circ}$  C.

(2) In a fruit cellar having a comparatively low and uniform

temperature ranging from 10° to 13° C.

(3) In the "dark room" of the botanical laboratory, which was quite dry and maintained at a temperature of 20° to 22° C.

(4) In the herbarium room on the fourth floor of the botanical laboratory. The air here was very dry and the mean temperature about the same as for No. 3, but with a much wider variation, reaching at times a maximum of 30° and a minumum of 10° C.

(5) In an incubator maintained at 30° to 32° C.

(6) In an incubator maintained at 37° to 40° C.

It will be observed that all of the preparations, except Nos. 1 and 4, were kept at temperatures which were quite uniform. The increase or decrease in the weight was determined at the expiration of 70 or 72 days by again carefully weighing the seed, after which germination tests were made. The results of the germination tests and the gain or loss in weight are given in Table X.

TABLE X.—Relation of moisture and temperature to vitality.

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Cabbage.	Perce	At the end of 36 hours,	14.4.4.4.4.4.4.4.4.4.4.4.4.4.4.4.4.4.4.
	ni serecontage of increase in weight while seeds were inclosed in bottles.		18888618392888243781988
дэ. Т	Amount of water put into each bottle.		8 <u>88888888888888888888888888888888888</u>
	Extremes of temper, ture for which seeds were subjected.		Degrave C. 10. 1.3. 10. 10. 1.3. 10. 10. 10. 10. 10. 10. 10. 10. 10. 10
	Place where seeds were stored.		Botanical laboratory Outdoors Fruit cellar Fruit cellar Fruit cellar Herbarium room Incubator Outdoors Outdoors Outdoors Dark room Herbarium room
		Labora- tory numbers,	Control. 1534 1534 1534 1535 1535 1535 1535 1539 1529 1529 1539 1529 1539 1529 1539 1539 1539 1539 1539 1539 1539 153

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1514. 1516. 1516. 1517. 1518.	1520 1521 1540 1541 1542 1544	1516.

a These bottles were simply closed with a cotton plug. bAir-dried seeds. Dried 30 days at 30° to 32° C, and then kept in open bottles until tests were made 40 days later.

The foregoing table, showing the conditions under which the seeds were kept, has been made quite complete. Aside from the final percentages of germination, the percentages of germination after a definite number of hours have likewise been given, the latter being better expressed as germinative energy. The germinative energy, as has been previously stated, is an important factor in determining the potential energy of a seed. This is quite clearly shown in many of the germination tests recorded in the above table. The preliminary results show a marked contrast as a result of the different kinds of treatment, while the final results reveal nothing more than the regular degree of variation usually met with in testing seeds. Of the five species of seeds, the onion has yielded the most striking variations in the earlier stages of germination. Take, for example, No. 1535, the sample that was kept in an open bottle in the fruit cellar. The moisture absorbed was sufficient to cause a chemical transformation, which injured the vitality of the seed and consequently caused a retardation in germination. No. 1539, the onion seed from the incubator maintained at a temperature of 37° to 40° C., germinated only 16.5 per cent in 77 hours, while the final percentage of germination was 95.5 per cent. Onion seeds Nos. 1532 and 1533 germinated in 77 hours 18.5 and 2.5 per cent respectively, while the final germination of the former was 93.5 per cent and of the latter 96 per cent. All of these tests gave final percentages of germination somewhat higher than the mean of the control samples. But the germination was considerably retarded, the control samples having germinated 29.5 per cent during the first 77 hours. These retardations in germination must be due to a lowering of vitality, as a more careful study of the table will show, and not to any excessive drying that may have taken place during the time of treatment. Numerous other examples are to be found in the table, some even more striking than those mentioned, but it is not deemed necessary that they all be pointed out and discussed here.

The table also shows the results of the various weighings made of all of the different samples which were kept in closed bottles. With but very few exceptions there was an increase in weight, which increase was quite marked in all cases where free water was introduced. The airdried seeds that were sealed in bottles without the introduction of free water all increased slightly in weight, with the exception of the peas, which showed a slight decrease in weight. It has been observed that the absolute loss in the weight of the peas was slightly greater than the total gain in the four other samples of seed. This, however, is not of sufficient uniformity throughout to fully justify the conclusion that cabbage, lettuce, onion, and tomato seed have a greater affinity for water than peas, and that the former robbed the latter of a portion of their water content. Yet a portion of the increased weight of the cabbage, lettuce, onion, and tomato seed is probably best accounted

for in that way. On the other hand, it is quite probable that a portion of the increase in weight was due to the results of intramolecular transformations and to the coexistent respiratory activities of the seed. The means of making these determinations are far from easy. Van Tieghem and G. Bonnier have shown" that seeds kept in sealed tubes in atmospheric air increased in weight during two years, but the increase was very small. In their experiments the peas which were in sealed tubes increased  $\frac{1}{790}$  of their original weight. A corresponding sample kept in the open air increased  $\frac{1}{72}$  of its original weight.

Nos. 1540 to 1545 in Table X show an increased weight in seeds when sealed in bottles for 70 days. These seeds were previously dried for 30 days at a temperature of 30 to 32°C. Disregarding the increase in weights as above given and the factors to which such increase may be attributed, it is quite evident that in all cases where water was added the increase in weight was due chiefly to the absorption of the water. The absolute increase was approximately the same as the weight of the water added.

The amount of water absorbed by different seeds varies greatly under identical conditions, depending largely upon the nature of the seed coats and the composition of the seed. The average increase in weight of the seeds used in these experiments was as follows: Onion, 6.27 per cent; pea, 5.51 per cent; cabbage, 4.12 per cent; lettuce, 3.99 per cent; tomato, 3.99 per cent. The loss in vitality of the corresponding samples was 28, 12, 23.7, 18.5, and 14.7 per cent, respectively. The relationship here is quite close, the amount of water absorbed being roughly proportional to the loss in vitality. The peas, however, afford an exception to this general statement. But it must be remembered that peas require a much larger percentage of moisture to start germination and are likewise capable of undergoing much wider variations than the other seeds in question. However, before a definite ratio can be established between the absorption of water and the loss in vitality, many other factors must be taken into consideration, such as the composition, water content, and duration of vitality of the seed under natural conditions.

Another interesting factor is shown in No. 1546 of Table X. These seeds were dried for 30 days at a temperature of 30 to 32° C., after which they were kept in an open bottle in the laboratory for 40 days. During the 30 days' drying the cabbage lost 2.41 per cent, lettuce 2.59 per cent, tomato 2.71 per cent, and the onion 3.47 per cent of moisture. These same seeds when exposed to the free air of the laboratory for 40 days never regained their original weight, the increase being as follows: Cabbage, 0.6 per cent; lettuce, 0.58 per cent; tomato, 1.56 per cent; onion, 0.89 per cent. The average quantity of water expelled was 2.79

a Bul. Soc. bot. France, 29: 25-29, 149-153, 1882.

per cent in 30 days, while the average increase in weight during the 40 days was only 0.91 per cent. These results show that if seeds are once carefully and thoroughly dried, they will remain so; that is, if kept in a comparatively dry room. This is an important factor in the preservation of vitality, as is borne out in the results of the germination tests. Later experiments were made with very similar results, and an analogous method of treatment promises to be of much value as a preliminary handling of seeds. It is not definitely known to what this stronger vitality is due, whether it be simply to the effect of the drying or to some process of chemical transformation which makes the seeds more viable. These results are now under consideration and will be reported at some future time.

The table also shows in a very striking degree the decrease in the number of germinable seeds with an increase in the moisture and temperature. The amount of moisture absorbed by the seeds, with a limited amount present in the bottles, was inversely proportional to the temperature. At the higher temperatures the inclosed air held a larger portion as water vapor; however, there was a greater deterioration in vitality. Where the seeds were kept outdoors at the low temperatures (-21.6° to 8.9° C.) of the winter months, no injury was apparent except where 3 cc. of water was added, and then only the onion seed was affected. This sample of seed had absorbed a quantity of water equal to 10.38 per cent of the original weight, which together with the original water content (6.41 per cent of the original sample) made 17.88 per cent of moisture in the seed. Practically the same results were obtained with the seeds kept in a fruit cellar at a temperature of 10° to 13° C. The samples of this series, in the open bottles, were also injured, as has been pointed out. With the samples that were stored in the dark room and in the herbarium room, the injury was more marked as a result of the higher temperature; but even here the seeds in the bottles which contained 0.5 cc. of free water deteriorated very little. The injury was confined to the onion seed, which showed a slight retardation in germination. Where 1 cc., 2 cc., and 3 cc. of water were added, vitality in some instances was likewise remarkably well preserved. The lettuce, tomato, and peas gave no indications of any deterioration save in the bottles containing 3 ec. of water. Here the lettuce and peas were permanently injured, while the tomato seeds suffered only sufficiently to cause a delay in the rapidity with which they germinated. The cabbage seed was retarded with 2 cc. and a lowering of the final percentage of germination with 3 cc. of water. The onion seed, being very sensitive to these unfavorable conditions, deteriorated very greatly, being practically worthless where 3 cc. of water were added. A brief study of the table will readily show that many seeds were killed at the still higher temperatures of 30 to 32 C. and 37 to 40 C. The onion seed was slightly injured even where

no water was added. However, a temperature of 40°C, is sufficient to injure many seeds, even though the liberated water be permitted to escape, as is shown in the tests of the onion, No. 1539 of the table. The greatest injury when air-dried seeds are sealed in bottles and then subjected to a higher temperature is due to the increased humidity of the confined air, as a result of the water liberated from the seeds.

At first glance some of the conditions given in the above table may seem to be extreme and far beyond any normal conditions that would be encountered in the ordinary handling of seeds. This may seem to be especially true with the seeds kept in the bottles with 3 cc. of water where the additional amount of moisture absorbed gave rise, in some of the seeds, to a water content of approximately 20 per cent. Yet this need not be thought of as an exception, for such extreme cases are often encountered in the commercial handling of seeds. During the process of curing even more drastic treatment is not infrequently met with. Pieters and Brown " have shown that the common methods employed in the harvesting and curing of Pou pratensis L. were such that the interior of the ricks reached a temperature of 130° to 140° F. (54.4° to 60° C.) in less than sixteen hours, at which temperature the vitality of the seed is greatly damaged and frequently entirely destroyed. The interior of one rick reached a temperature of 148° F. (64.4° C.) in twenty hours, and the vitality had decreased from 91 per cent to 3 per cent, as shown by the germination of samples taken simultaneously from the top and from the inside of the same rick.

On the other hand, the extreme cases need not be considered. Take, for example, the onion seed that was sealed in a bottle with 1 cc. of water and maintained at a temperature of 37 to 40 °C. The increase in weight due to the water absorbed was 3.91 per cent, thus giving a moisture content of 11.2 per cent and a complete destruction of vitality. The cabbage seed, kept in the same bottle, had absorbed a quantity of water equivalent to 2.35 per cent of its original weight, which, with the 5.90 per cent contained in the original sample, gave 8.25 per cent of water. This sample of seed germinated only 11 per cent, having thus no economic value. In neither of these samples was the amount of water present in the seeds greater than that ordinarily found in commercial samples. Moreover, the temperature was much below that frequently met with in places where seeds are offered for sale and likewise well within the limits of the maximum temperature of our summer months, especially in the Southern States. Take, by way of comparison, the maximum temperatures of some of the places at which seeds were stored to determine the effect of climate on vitality, as shown in another part of this paper. During

a Bulletin 19, Bureau of Plant Industry, U. S. Department of Agriculture, 1902.

the summer of 1900 the maximum temperature at Wagoner, Ind. T., was 107° F. (41.1° C.), while that of Lake City, Fla., was 103° F. (39.5 C.). If these points are kept in mind, it is not at all surprising to find that seeds lose their vitality within a few weeks or months in warm, moist climates.

In order to make the above facts more clear the preceding table has been summarized and is presented in the following condensed form, showing the relation of the water content of the seed to vitality:

Table XI.—Marked deterioration in vitality with an increase in the quantity of the water content of seeds.

How preparations were made.	Amount of water introduced into the bottles.	Average increase in weight as a result of the greater water content.		Average germination.
Control sample	cc.	Per cent.	Per cent. 6,07	Per cent. 93.3
Closed bottles, sealed with paraffin	Water expelled.	0.06	a 2.77	a 93. 9
Do	None.	.08	6, 55	94.0
Do	0.5	1.75	8.31	91.7
Do	1.0	3.24	9.91	83, 3
Do	2.0	5, 91	12.75	67.5
Do	3.0	8.13	15.10	58, 6

a Peas not included in this set.

Numerous other results of a similar character might be cited, but it hardly seems necessary at this time, since there can be no doubt that moisture is the prime factor in causing the premature destruction of vitality in seeds in the usual conditions of storage. Why they lose their vitality as a result of the unfavorable conditions is quite a different question, and has to do with the very complex composition of the seed.

# A COMPARISON OF METHODS OF STORING AND SHIPPING SEEDS IN ORDER TO PROTECT THEM FROM MOISTURE AND CONSEQUENTLY TO INSURE A BETTER PRESERVATION OF VITALITY.

#### SUGGESTIONS OF EARLIER INVESTIGATORS.

As early as 1832, Aug. Pyr. De Candolle" wrote a chapter on the conservation of seeds, in which he said that if seeds be protected from moisture, heat, and oxygen, which are necessary for germination, their vitality will be much prolonged; moreover, that if seeds are buried sufficiently deep in the soil, so that they are protected at all times from the very great influence of oxygen and moisture, their vitality will be preserved for a much longer period.

# Giglioli<sup>a</sup> goes so far as to say:

There is no reason for denying the possibility of the retention of vitality in seeds preserved during many centuries, such as the Mummy wheat and seeds from Pompeii and Herculaneum, provided that these seeds have been preserved from the beginning in conditions unfavorable to chemical change. \* \* \* The original dryness of the seeds and their preservation from moisture or moist air must be the very first conditions for a latent secular vitality.

Some of the earliest suggestions for storing seeds in quantity were made by Clément and Fazy-Pasteur, and were reported by Aug. Pyr. De Candolle in his Physiologie Végétale. Clément suggested the use of large cast-iron receptacles, made impervious to air and water, the well-dried seeds to be poured in through an opening at the top, after which the opening should be hermetically sealed and the seeds withdrawn through an iron pipe and stopcock at the bottom of the tank. The scheme of Fazy-Pasteur was to store seeds in wooden boxes well covered with tar. This method was especially applicable to small quantities of seeds, and was used to a limited extent at that time, but, so far as has been ascertained, it has long since been discarded. keeping of seeds in large iron tanks, as suggested by Clément, has never been practiced to any extent. It seems quite possible, however, that the present "tank" grain elevator, now so universally used, might readily be modified in such a way as to make the method suggested by Clément quite practicable.

#### THE NECESSITY FOR THOROUGHLY CURING AND DRYING SEEDS.

In addition to being well matured and carefully harvested, seeds should be thoroughly cured and dried before being put into the storage bins. Much better results would be obtained if such seeds were artificially dried for several days in a current of dry air at a temperature not to exceed 35° C. With this method of drying, from 2 to 4 per cent of the moisture usually present in air-dried seeds is expelled. The accompanying contraction of the seed coats makes them more impervious to the action of moisture, and consequently the seeds are better prepared for storing and shipping. Experiments made with cabbage, lettuce, onion, and tomato seeds gave results as follows: The average loss in weight of the air-dried seeds, after an additional drying of 30 days at a temperature of 30° to 32° C. was 2.79 per cent. Yet these same seeds, when kept for 40 days in the laboratory, reabsorbed only an average of 0.91 per cent of moisture. Like quantities from the original sample gave only the slight variations ordinarily met with, due to the humidity of the atmosphere. Thus seeds, when once carefully and thoroughly dried, will not regain their original weight, provided they be kept in a dry room.

#### CHARACTER OF THE SEED WAREHOUSE OR STORAGE ROOM.

Another important factor in the storing of seeds is the character of the seed warehouse or storage room. The first point to be considered is dryness. Such houses should be kept as dry as possible, which can be accomplished either by means of artificial heat or by the use of strong drying agents, or better still, by both. True, if the seed warehouse be located in a section having a dry climate, this difficulty is at once largely overcome. But in many cases such a location is impraeticable or even impossible, and other means must be resorted to. As a matter of fact, most large seed warehouses are not heated and a great loss in vitality inevitably follows; but each seedsman must determine for himself whether or not this loss is sufficiently great to justify the expense of heating such a storage room.

Experiments carried on during the progress of this work have shown some very marked differences in favor of seeds stored in rooms artificially heated. The averages of the thirteen samples of seeds from the eight places at which they were stored show a difference in the loss of vitality of 9.87 per cent. Those kept in rooms that were artificially heated during a greater portion of the time deteriorated 25.91 per cent, while those stored in rooms not so heated deteriorated 35.78 per cent. The loss here given for seeds stored in dry rooms is greater than such conditions warrant, owing to the very unfavorable conditions at Mobile, Ala., and Baton Rouge, La. At Lake City, Fla., the relative percentages of deterioration were 29.42 and 16.27 for the unheated and heated rooms, respectively; at Auburn, Ala., 33.90 and 10.34 per cent, and at Durham, N. H., 39.58 and 3.57 per cent, respectively. Unfortunately these experiments were not made with this definite point in view, and the results are not entirely satisfactory, as no records were made of the temperatures and humidities.

### THE VALUE OF GOOD SEED TO THE MARKET GARDENER.

This work was undertaken chiefly for the purpose of finding some improved methods of shipping and storing seeds in small packages, wherein their vitality might be better preserved. The rapid deterioration in vitality causes great losses to gardeners living in districts where the climatic conditions bring about the premature destruction of vitality in seeds. In many cases the seeds are practically worthless or altogether fail to germinate after a few weeks' exposure. The loss in such cases is not in the greater quantity of seed required, but the retardation or complete failure of the germination often means delay, making the difference between success and failure in the desired crop. Seed of low vitality is even worse than dead seed. With the latter the difficulty is soon discovered, while with the former, although the seed will germinate, the seedlings are not sufficiently vigorous to develop

into strong and healthy plants. True, most enterprising gardeners usually have vitality tests made immediately preparatory to planting, but this is not always convenient, and they rely on the results of tests made at some earlier date. In such cases it quite frequently happens that they accept the results of tests made several weeks earlier. With many seeds this will suffice, yet there are many others that will deteriorate very materially within a few weeks or even within a few days in such unfavorable climates as exist, for example, near the Gulf of Mexico. In a letter dated January 15, 1903, Mr. J. Steckler, of New Orleans, La., wrote as follows concerning the vitality of seeds:

Some seeds are not worth being planted after being here three months. This is especially true of cauliflower seed. We have made repeated tests and this seed after remaining here 90 days was worthless and had to be thrown away.

#### SHIPPING SEEDS IN CHARCOAL, MOSS, ETC.

Bornemann<sup>a</sup> made some experiments with seeds of *Victoria regia* and *Euryale ferox*, in which he found that when packed in powdered charcoal they soon lost their vitality, but when packed in powdered chalk slightly better results were obtained. On the other hand, Dammer<sup>b</sup> recommends powdered charcoal as a method of packing for seeds that lose their vitality during shipment, especially the seeds of palms and a number of the conifers.

Charcoal is undoubtedly much better than moist earth or moss, which are frequently used, the latter affording abundant opportunities for the development of molds and bacteria during transit. Some such method as moist charcoal is necessary in case of seeds which lose their vitality on becoming dry. Numerous other reports have been published from time to time concerning the shipping of seeds of aquatic plants, as well as those of low vitality, but they need not be discussed further at this time.

#### NATURE OF THE EXPERIMENTS.

Aside from some popular accounts and miscellaneous suggestions, but little has been done toward finding improved methods of shipping and storing seeds of our common plants of the garden and field. Accordingly, in February, 1900, a series of experiments was undertaken to determine some of these factors, in which three questions were considered: (1) How may small quantities of seeds be put up so as to retain a maximum germinative energy for the greatest length of time? (2) What immediate external conditions are best suited for the longevity of seeds? (3) What part do climatic conditions play in affecting the life of seeds?

<sup>&</sup>lt;sup>a</sup> Gartenflora, 35. Jahrg., 1886, pp. 532-534.

<sup>&</sup>lt;sup>b</sup> Ztschr. trop. Landw., Bd. I, 1897, No. 2.

In order to answer the first question, duplicate samples of the various kinds of seeds were put up in double manila coin envelopes, as described on page 14. Likewise, duplicate samples were put up in small bottles, the bottles being closed with good cork stoppers. Some of the bottles were filled with seed, while others were only partly full. In some cases there was a surplus air space five times as great as the volume of the inclosed seeds. This space, however, had no bearing on the vitality of the seeds as far as could be determined.

In order to determine what immediate external conditions play an important part in the destruction of vitality, samples of seed, prepared as above described, were stored in different places." At each place they were subjected to three different conditions of storage, which, for convenience, have been designated as "trade conditions," "dry room," and "basement," as described on page 14. In addition to these three methods of storage, numerous other conditions were tried in and near the laboratory; such as in incubators at increased temperatures and with varying degrees of moisture, in cold storage, in greenhouses, and in various gases, in vacuo, in liquids, etc.

The third question, "What part do climatic conditions play in affecting the life of seeds?" has been answered for the most part in a discussion on the effect of climate on vitality, page 13. In fact, the seeds in the envelopes kept under trade conditions were the same in both cases, being used here simply as a means for comparing the vitality of seeds when stored in paper packages and in bottles, as well as to show the relative merits of trade conditions, dry rooms, and basements as storage places for seeds.

# DISPOSITION OF THE SAMPLES.

A more definite description of the treatment given the seeds in the various places may be summed up as follows:

San Juan, P. R.—The seeds were sent to San Juan on February 9, 1900, and were returned on June 20, 1900, after a lapse of 131 days. At San Juan the seeds were stored under trade conditions only, and the various packages were not removed from the original box in which they were sent. While in San Juan the box containing the seeds was kept in a room well exposed to climatic influences, being protected only from the direct rays of the sun and from rain.

<sup>&</sup>quot;San Juan, P. R.; Lake City, Fla.; Mobile, Ala.; Auburn, Ala.; Baton Rouge, La.; Wagoner, Ind. T.; Durham, N. H., and Ann Arbor, Mich.

<sup>&</sup>lt;sup>b</sup> The places of storage represented by trade conditions have already been described for each of the localities, but it seems advisable to rewrite the descriptions here so that they may be more readily compared with the dry room and basement conditions.

<sup>&</sup>lt;sup>c</sup>The exact time that the seeds remained at San Juan was much less than 131 days, the time of transportation being included, as has been done for the other places.

Lake City, Fla.—The seeds were sent to Lake City on February 9, 1900. The first complete set was returned on June 18, after 129 days. The second complete set was returned October 1, after 234 days. The "trade conditions" at Lake City were supplied by keeping the seeds in a small, one-story frame building, the doors of which were open the greater part of the time. This building was not heated, and the seeds were stored approximately 5 feet from the ground. "Dry room" conditions were those of a storage room on the fourth floor of the main building of the Florida Agricultural College. The third set was kept in a small bulletin room in the basement of the same building.

Mobile, Ala.—The seeds were sent to Mobile on February 17, 1900. One set was received in return on July 7, after 180 days. The other set was received on November 6, after 262 days. The "trade conditions" in this case consisted of a comparatively open attic in a one-story frame dwelling. The set in a "dry room" was kept in a kitchen on a shelf 5 feet from the floor, and not more than 6 feet distant from the stove. Here they were subjected to the action of artificial heat throughout the entire period. The seeds under "basement" conditions were kept in a small cellar, which during the season of 1900 was very moist.

Auburn, Ala.—The seeds were sent to Auburn on February 17, 1900. The first complete set was received in return on May 30, the second on November 19 of the same year, or after 102 and 275 days, respectively. "Trade conditions" consisted of an office room connected with a greenhouse, with the doors frequently standing open; "dry room" conditions were obtained in the culture room of the biological laboratory on the third floor of the main huilding of the Alabama Polytechnic Institute, "basement" conditions being found in the basement of the same building, a comparatively cool situation, yet with a relatively high degree of humidity.

Baton Ronge, La.—The seeds were sent to Baton Rouge on February 17, 1900. On June 18 the first complete set was received in return. The second set remained until October 22, making the time of absence 121 days for the first and 247 for the second set. "Trade conditions" at Baton Rouge were furnished by keeping the seeds throughout the entire time of the experiment on shelves in a grocery store, the doors of which were not closed except at night. These conditions were thus identical with those to which seeds are subjected when placed on sale in small stores. The "dry room" was a class room on the second floor in one of the college buildings. A storeroom in the basement of a private residence, having two sides walled with brick, furnished "basement" conditions.

a Presumably these were in a dry place, but further evidence showed that the presumption was erroneous. The vapors arising while cooking was being done on the stove gave rise to conditions very detrimental to a prolonged life of the seeds.

<sup>25037-</sup>No. 58-04-4

Wagoner, Ind. T.—The seeds were sent to Wagoner on February 17, 1900. The first series was received in return on June 23, after 126 days; the second set was returned after 238 days, on October 13, 1900. The sets for "trade conditions" were kept in a drug store, on a counter near an open door. The "dry room" was a sleeping room on the first floor of the same building, while "basement" conditions were supplied by keeping the seeds in a large depository vault in a bank.

Durham, N. H.—The two sets of seeds were sent to Durham on February 17, 1900, and were returned on July 14 and October 20, after 117 and 231 days, respectively. The seeds under "trade conditions" were kept over a door at the entrance of one of the college buildings. The door opened into a hall, which led into office rooms, the chemical laboratory, and the basement. An office room on the first floor of the same building supplied "dry room" conditions. The seeds were located well toward the top of the room, which was heated with steam and remained quite dry at all times. The "basement" conditions were found in a storage room in one corner of the basement of the same building.

Ann Arbor, Mich.—The set of samples placed under "trade conditions" was kept in the botanical laboratory, being moved about from time to time in order to supply the necessary variations to an herbarium room, to an open window, and to an attic. From February 18, 1900, until May 12, 1900, the set of seeds under "dry room" conditions was stored in a furnace room. The seeds were only a few feet from the furnace and were always quite dry and warm: The maximum temperature recorded was 43° C., with a mean of 38° during cold weather, and of 30° C. during milder weather. On May 12 this set of seeds was transferred to the herbarium room on the fourth floor of the botanical laboratory, where they remained until vitality tests were made. "Basement" conditions were found in a fruit cellar, having two outside walls and a temperature fluctuating between 10° and 13° C.

These packages and bottles were all securely packed in new cedar boxes from which they were not removed until after their return to the laboratory.

#### RESULTS OF THE GERMINATION TESTS.

After receipt of the seeds, germination tests were made as rapidly as possible, the results of which are given in the tabulations which follow. Likewise, in each case is shown the vitality of the control sample. Furthermore, a summary of each table is given, showing the average percentages of germination of the seed from the various places for the first and second tests, respectively. From these results the average percentage of loss in vitality has been calculated, reckoning the germination of the control sample as a standard. It is thus a very simple matter to compare the relative merits of the different methods of storing and the rôle they play in promoting the longevity of seeds.

Table XII.—Percentage of germination of beans subjected to various conditions of storage in different localities.

[Germination of control sample: First test, 98.7 per cent; second test, 98.7 per cent.]

				Perce	ntage of	germin	ation.	
Place of storage.	Order of tests.	Num- ber of days in		e con-	Dry rooms.		Basements.	
		storage.	Envel- opes.	Bottles.	Envel- opes.	Bottles.	Envel- opes.	Bottles.
Lake City, Fla Do	First Second.	129 234	98 84	98 98	98 96	98 98	86 0	98 100
Auburn, Ala	First Second.	102 275	98 56	97.5 98	100 91	100 98	97. 9 66	97.5 100
Mobile, Ala	First Second.	140 262	58 0	96 90	82 0	100 98	0	100 98
Baton Rouge, La	First Second.	121 247	96 60	100 96	92 28	100 100	54 0	98 98
San Juan, P. R.	First Second.	131	100 96	100 98				
Wagoner, Ind. T	First Second.	126 238	96 82	96 100	98	100 100	100 84	98 98
Durham, N. H	First Second.	147 251	100 78	100 . 96	100 98	98 96	100 92	100 98
Ann Arbor, Mich	First Second.		98 100	84 100	98 100	84 91.5	98 92	92 100
Average percentage of ger-	[First	128	93	96.44	95. 43	97.14	66, 99	97.61
mination.	Second .	251	69.50	97	69.33	97.36	55, 66	98.86
Average percentage of gain	[First	128	5.78	2.29	3.31	1.58	32, 13	1.06
or loss in vitality.	Second .	251	29.59	. 1.72	29.76	1.36	43, 61	+0.10

The beans at Mobile were seriously affected under all conditions except when put up in bottles and thus protected from the moist atmosphere. Those kept in bottles under "trade conditions" deteriorated to 90 per cent, but the result of the first test of the same series indicates that some moisture passed through the cork and that the seeds were injured in that way.

At Baton Rouge the beans retained their vitality somewhat better; but even here all those from the envelopes were practically worthless after 247 days, for beans that germinate only 60 per cent are of no value for planting.

The "trade conditions" at Auburn, Ala., and Durham, N. II., were also very unfavorable to the prolonged vitality of the beans. At Wagoner, Ind. T., San Juan, P. R., and Lake City, Fla., there was a marked deterioration, yet not sufficiently great during the time given to render them worthless for planting. However, it is quite evident that beans subjected to such conditions of storage would not be fit for planting the second season.

A summary of the table shows that the vitality of the beans when kept in bottles and subjected to either of the three conditions was not interfered with. The averages show a variation of less than 2 per cent. With those kept in paper packages the results were quite different, the advantage being slightly in favor of the "trade conditions." The loss in vitality was 29.59, 29.76, and 43.61 per cent, respectively, for "trade conditions," "dry rooms," and "basements."

Table XIII.—Percentage of germination of peas subjected to various conditions of storage in different localities.

ALTERNATION AND THE PROPERTY OF THE PROPERTY O		Num- ber of days in storage.	Percentage of germination.							
Place of storage.	Order of tests.		Trade tio		Dry r	ooms.	Basen	nents.		
			Envel- opes.	Bottles.	Envel- opes.	Bottles.	Envel- opes.	Bottles.		
Lake City, Fla Do	First Second.	129 234	96 86	97. 9 98	94 92	94 92	96 6	98 98		
Auburn, Ala Do	First Second.	102 275	93.3 97.9	94 . 91	87.8 90	97.8 96	93. 9 86	94 98		
Mobile, Ala	First	140 262	$69.2 \\ 44$	92 100	88 42	96 96	10.2	98 98		
Baton Rouge, La	First Second.	121 247	94 80	92 88	94 70	90 98	90 0	98 98		
San Juan, P. R	First	131	91 98	100 98						
Wagoner, Ind. T	First	126 238	98 80	90 92	96	92 96	90 88	88 92		
Durham, N. H	First Second.	147 251	98 94	94 98	100 94.7	98 96	94 98	98 90		
Ann Arbor, Mich	First Second .		90 98	94 94	94 94	72 92	96 86	94 100		
Average percentage of ger-	First	128	91.56	94.24	93.4	91.41	81.44	95.43		
mination.	Second .	251	84.74	95.25	80.45	95.14	60, 66	96.28		
Average percentage of gain	[First	128	3,92	1.12	1.99	4.08	14.55	+0.14		
or loss in vitality.	Second .	251	11, 45	0.47	15.94	0.58	36.62	+0.60		

The peas retained their vitality much better than the beans. However, the greatest loss in both peas and beans was in the envelopes at Mobile and Baton Rouge. Some of the samples from the envelopes germinated fully as well or even better than the control, but the general averages of the second tests for all of the localities show a loss of 11.45 per cent in "trade conditions," 15.94 per cent in "dry rooms," and 36.63 per cent in "basements." The beans under identical conditions lost 29.59, 29.76, and 43.61 per cent, respectively.

The seeds kept in bottles deviated but very little from the standard of the control.

Table XIV.—Percentage of germination of cabbage subjected to various conditions of storage in different localities.

[Germination of control sample: First test, 92.7 per cent; second test, 92.4 per cent.]

		Num- ber of days in storage.	Percentage of germination.							
Place of storage.				condi- ns,	Dry rooms.		Basements.			
			Envel- opes.	Bottles.	Envel- opes.	Bottles.	Envel- opes.	Bottles.		
Lake City, Fla	First Second.	129 234	89.5 63.5	92. 5 89. 5	89.5 81.5	94 89.5	86.5 14.5	90.5 94.5		
Auburn, Ala Do	First Second.	· 102 275	91 61, 5	90.5 90	89.5 90	81 89	92 60	91 85, 5		
Mobile, Ala Do	First Second.	$\frac{140}{262}$	64.5 17	93.5 87.5	58. 5 5	96 95	58, 5	92.5 94		
Baton Rouge, La	First Second.	121 247	88.5 25.5	93 90, 5	90.5 11.5	91 86	79.5 0.5	94 90.5		
San Juan, P. R	First Second.	131	82 76. 2	95.5 89						
Wagoner, Ind. T	First Second.	126 238	83.5 70.5	93 91, 5	91	95, 5 92, 5	88.5 76.5	97.5 89		
Durham, N. H	First Second.	147 251	93 12	97.5 92.5	89 93	96 95, 5	95.5 92.5	94.5 96.5		
Ann Arbor, Mich			96 91	92 94	94 88	90.5 82	89.5 76	94, 5 95, 5		
Average percentage of ger-	First	128	86	93.47	86.43	92	84.29	93.5		
mination.	Second .	251	52.15	90.56	61.5	89.93	53, 33	92, 21		
Average percentage of gain or loss in vitality.	First Second.	128 251	7.23 43.56	+0.83 1.94	6.77 33.44	0.86 2.67	9.07 42.29	+0.86 0.22		

Table XIV shows that the cabbage, like the peas, was injured to a less degree at Mobile and Baton Rouge than the beans, but even the cabbage seed kept in the paper packages in these cities were all but killed. The average degree of injury, however, was greater in the cabbage than in the beans. In a majority of cases there was more or less deterioration in the case of this seed kept in the envelopes. Aside from those already mentioned, the trade conditions at Durham, N. H., and the basement at Lake City, Fla., should be expressly noted.

The seeds kept in the bottles deviated but little from the control, while those kept in paper packages germinated only 52.15, 61.50, and 53.33 per cent for the trade conditions, dry room, and basement—equivalent to a loss in vitality of 43.56, 33.44, and 42.29 per cent, respectively.

Table XV.—Percentage of germination of radish subjected to various conditions of storage in different localities.

[Germination of control sample: First test, 83.6 per cent; second test, 78.8 per cent.]

		Num- ber of days in	Percentage of germination.						
Place of storage.	Order of tests.			condi- ns.	Dry rooms.		Basements.		
		storage.	Envel- opes.	Bottles.	Envel- opes.	Bottles.	Envel- opes.	Bottles.	
Lake City, Fla	First Second.	129 234	79 58, 5	78. 5 64	84.5 67.5	75 71.5	66 48.5	83 67	
Auburn, Ala		102 275	75.5 63	85   72,5	85, 5 66	80.5 73.5	86, 5 60, 5	85.5 76.5	
Mobile, Ala		140 262	58.5 51	81 71.5	56.5 49	81 70	75	76 72	
Baton Rouge, La	First	121 247	77.5 55.5	85, 5 69, 5	73.5 49.5	78.5 71.5	61, 5 51, 5	78, 5 75	
San Juan, P. R	First Second.	131	64 62	81, 5 73, 5					
Wagoner, Ind. T	First Second .	126 238	77.5 60,5	80, 5 75, 5	79	81 77	80, 5 63	86.5 70.5	
Durham, N. H		147 251	80, 6 59, 5	75, 5 81, 5	76.5 74.5	85 85	81 68	74 79	
Ann Arbor, Mich	First Second.		82.5 77.5	85   80, 5	82.5 79.5	79. 5 57. 5	78 62, 5	82. 9 78. 5	
Average percentage of germination,	First   Second.	128 251	74, 39 60, 94	81,56 73,56	76, 86 64, 33	80, 5 72, 71	75, 5 59	80, 91 74, 07	
Average percentage of loss in vitality.	First Second.	128 251	11.02 22.67	2, 44 6, 65	8.07 18.37	3.71 7.73	9, 67 25, 13	3. 22 6	

The results of the tests of the radish seed are very similar to those of the cabbage; the latter, however, showed a greater loss in vitality. As shown by the second tests, the average percentages of deterioration of the cabbage seed which was kept in the envelopes were as follows: Trade conditions, 43.56 per cent; dry room, 33.44 per cent; basement, 42.29 per cent, while the loss in vitality of the radish was only 22.67, 18.37, and 25.13 per cent, respectively.

Table XVI.—Percentage of germination of carrot subjected to various conditions of storage in different localities.

[Germination of control sample: First test, 83.3 per cent; second test, 82 per cent.]

			Percentage of germination.						
Place of storage.	Order of tests.	Num- ber of days in	tio	condi- ns.	Dry rooms.		Basements.		
		storage.	Envel- opes.	Bottles.	Envel- opes.	Bottles.	Envel- opes.	Bottles.	
Lake City, Fla Do	First Second.		76.5 43.5	83 80, 5	78 67.5	78.5 78.5	73 3	77.5 81.5	
Auburn, Ala	First Second.	102 275	84.5 36	82 76.5	83 72.5 ,	86 76. 5	86.5 47.5	86, 5 82, 5	
Mobile, Ala	First Second.		59 8.5	87.5 86	51.5 .5	83.5 69	20,5	87 78	
Baton Rouge, La	First Second.	121 247	74.3 25	82.3 72.5	75.1 16.5	86.8 52.5	57.3 0	82, 3 39	
San Juan, P. R.	First Second.	131	71.5 48.5	82.5 86.5					
Wagoner, Ind. T	First Second.	126 238	81.5 49	82 81.5	77,5	81 81	77.5 45.5	87.5 84	
Durham, N. H	First	147 251	78 2	82.5 85.5	84 87.5	85.5 85.5	83.5 72	82.5 87.5	
Ann Arbor, Mich			76 86	79 78	83 78.5	75.5 80	78 58, 5	83.5 71	
Average percentage of ger-	1	128	75.16	82.6	76.01	82.4	68, 04	83, 83	
mination.	(Second.	251	37.31	80.87	53,83	74.71	37.75	75, 21	
Average percentage of gain or loss in vitality.	First	128 251	9,72 54,5	0.84 1.38	8, 75 34, 35	1.08 8.89	18, 32 53, 96	+0.63	
						2100			

Table XVI shows results very similar to those of Table XV, except that the carrot was affected slightly more than the cabbage. There was also a greater falling off in the case of the seeds kept in the bottles in dry rooms and basements. The reason for this is not very clear. Apparently it was due to some local conditions, inasmuch as it was confined chiefly to the bottles kept at Mobile and Baton Rouge. The average results of the germination tests of the seeds kept in packages are quite low for the carrots. Seed from trade conditions germinated 37.31 per cent, from basements 37.67 per cent, and from dry rooms 53.83 per cent, with a loss in vitality of 54.5, 54.06, and 34.36 per cent, respectively. Under similar conditions the cabbage lost in vitality 43.56, 42.28, and 33.45 per cent, respectively.

Table XVII.—Percentage of germination of "A" sweet corn subjected to various conditions of storage in different localities.

[Germination of control sample: First test, 92.7 per cent; second test, 92.4 per cent.]

		Num- ber of days in	Percentage of germination.						
Place of storage.	Order of tests.		Trade condi- tions.		Dry rooms.		Basements.		
		storage.	Envel- opes.	Bottles.	Envel- opes.	Bottles.	Envel- opes.	Bottles.	
Lake City, Fla	First Second.	129 234	94 92	96 100	94 96	92 90	88 54, 5	98 100	
Auburn, Ala	First Second.	102 275	96 88	98 98	94 91	98 90	100 80	92 100	
Mobile, Ala	First Second .	140 262	80 20	100 96	80 26	96 100	94.1	96 96	
Baton Rouge, La	First Second .	121 247	96 88	94 96	96 88	88 96	86 14	100 100	
San Juan, P. R	First Second .	131	96 92	94 94					
Wagoner, Ind. T	First Second .		96 90	98 96	94	96 96	96 92	96 94	
Durham, N. H	First Second.	147 251	100 96	92 96	95. 9 96	90 96	100 100	96 98	
Ann Arbor, Mich	First Second.		100 98	86 98	94 100	89 96	100 92	96 98	
Average percentage of ger-	First		94.75	94.75	92.56	94.14	94.87	96.29	
mination.	[Second .	251	83	96.75	83, 33	94.86	72,08	98	
Average percentage of gain	First		+2.21 $10.11$	+2.21	0.15 9.81	+0.01  +2.66	$\begin{vmatrix} -2.34 \\ 22 \end{vmatrix}$	+3.87	
or loss in vitality.	(second .	251	10.11	+4.71	9.81	+2.00	1 22		

Table XVIII.—Percentage of germination of "B" sweet corn subjected to various conditions of storage in different localities.

[Germination of control sample: First test, 89.3 per cent; second test, 88.5 per cent.]

				Perce	ntage of	germin	ation.	-
Place of storage.	Order of tests.	Num- ber of days in storage.	Trade condi- tions.		Dry rooms.		Basements.	
			Envel- opes.	Bottles.	Envel- opes.	Bottles.	Envel- opes.	Bottles.
Lake City, Fla	First Second .	129 234	86 77.1	60	90 64	38 0	76 30	46 0
Auburn, Ala	First Second .	102 275	88 62	92 56	86 82	86 38	86 82	84 89, 6
Mobile, Ala	First Second .	140 262	48 12	81.2 52	60 16	87.5 54	75	86 76
Baton Rouge, La	First Second .	121 247	80 54, 2	82 36	84 66	91 46	61 4,5	88 61. 2
San Juan, P. R	First Second .	131	72 78	72 71.7				
Wagoner, Ind. T	First Second .	126 238	70 78	82 76	90	88 88	84 88	84 76
Durham, N. H	First Second .	147 251	89.3 82	69.5 91.8	84.2	83.6 88	80 76	80 88
Ann Arbor, Mich	First Second .		92 80	88 92	88 86	48 22	88 82	96 88
Average percentage of germination.	First	128 251	78.16 65.41	78.31 59.70	83, 17 66, 33	75.01 48	79 60, 41	80, 55 68, 40
Average percentage of loss	[First	128	12, 47	12.31	6.87	16	11.51	9, 80
in vitality.	Second .	251	26, 09	32, 55	25, 06	45.76	31.74	22.71

Tables XVII and XVIII have been considered together, since both have to do with the same variety of sweet corn. The difference in the quality of these two samples was quite marked when the seed was received. Germination tests were made January 30, 1900, and showed 94 per cent for the "A" and 88 per cent for the "B" corn. November, 1900, samples of seed from the same original packages were tested, giving a germination of 92.4 per cent and 88.5 per cent for the "A" and "B" samples, respectively, as shown in the controls of the above tables. Thus, when two grades of corn are subjected to favorable conditions of storage, both are well preserved; but when subjected to unfavorable conditions, the one of poorer quality is much more susceptible to injury. The "A" sample which was stored in envelopes in trade conditions lost 10.11 per cent, as compared with 26.9 per cent for the "B" sample. The "A" sample which was stored in dry rooms lost only 9.81 per cent, while the "B" sample lost 25.06 per cent. In basements, the "A" sample lost 23 per cent and the "B" sample 31.74 per cent. In both samples the corn in the packages stored in the basement at Mobile was so badly molded at the time the second tests were made that they have been omitted from the table.

The most interesting feature in comparing the results of these two samples is found in the seed which was stored in the bottles. The average results of the "A" samples show a much higher percentage of germination for those from the bottles than the control, while the averages for the "B" sample were much lower than the corresponding controls. The average germination of the "B" sample from the bottles was 59.7 per cent for the trade conditions, 48 per cent for dry rooms, and 68.4 per cent for basements, or a loss in vitality of 32.55, 45.76, and 22.71 per cent, respectively. This difference was due to two causes, first, a difference in the quality of the seed at the beginning of the experiment, and, secondly, the larger amount of water in the second sample, "B." The greater quantity of water present in the seed gave rise to a more humid atmosphere after the seeds were put into the bottles, especially when subjected to higher temperatures than those in which the seeds had been previously stored. This is an important factor always to be borne in mind when seeds are put up in closed receptacles; they must be well dried if vitality is to be preserved.

Table XIX.—Percentage of germination of lettuce subjected to various conditions of storage in different localities.

[Germination of control sample: First test, 81.6 per cent; second test, 92.3 per cent.]

				Perce	ntage o	f germin	ation.	
Place of storage.	Order of tests.	Num- ber of days in		ade itions.	Dry rooms.		Basements.	
		storage.	Envel- opes.	Bottles.	Envel- opes.	Bottles.	Envel- opes.	Bottles.
Lake City, FlaDo	First Second.		87 85	81 92	81 92, 5	76.5 90	68 43, 5	77 95, 5
Auburn, Ala	First Second.	102 275	86.5 86	85.5 90.5	88.5 90.5	84.5 91	84: 5 83, 5	88, 5 90
Mobile, Ala	First Second.	140 262	63 20	78 88.5	58 31	87.5 90.5	1.5	83 91.5
Baton Rouge, La	First Second.		82.5 84.5	81.5 93.5	79 74.5	78.5 87.5	70.5	76 92.5
San Juan, P. R	First Second .	131	79 83, 5	87.5 89				
Wagoner, Ind. T	First		78 82	76 92.5	80	82   94	81 87.5	76.5 89
Durham, N. H	First		82.5 88.5	80.25	83, 25 92	77.5	80 90, 5	75.2 90.5
Ann Arbor, Mich	First		82 92. 5	68.5	84.5 89.5	81.5	78.5 88	72 91, 5
Average percentage of germination,	First   Second .		80, 06 77, 75	80.15 91.12	79.18 78.33	81.14 90.93	66, 28 65, 58	78.31 90.78
Average percentage of loss in vitality:	First  Second.	128 251	1.89 15.76	1.77	2. 97 15. 14	.56 1.49	18.78 28.95	4. 03 1. 65

The lettuce has shown no very marked deviation from the controls, save the seeds from the packages kept at Mobile, and those which were stored in basements in envelopes at Baton Rouge and Lake City. The average results of the second series of tests show a similar loss in vitality of all of the seeds from the envelopes. The samples of seed from the bottles germinated practically as well as the controls. The results of the first series of tests are not entirely satisfactory, none of the tests having gone to standard. The low germination of the lettuce in this series was due to inability to properly control the temperature in the germination of lettuce seed is 20°C, while in this first series the germination tests were unavoidably made at 26° to 27.5°C. Nevertheless, this seeming objection is of little consequence, since all of the results are directly comparable with the control.

Table XX.—Percentage of germination of onion subjected to various conditions of storage in different localities.

[Germination of control sample: First test, 95.8 per cent; second test, 97 per cent.]

M. 40 W.		Num- ber of days in storage.	Percentage of germination.							
Place of storage.	Order of tests.			condi- ns.	Dry rooms.		Basements.			
			Envel- opes.	Bottles.	Envel- opes.	Bottles.	Envel- opes,	Bottles.		
Lake City, Fla	First Second.	129 234	95 16.5	95 95. 5	95, 5 79	95 96	80 0	97. 5 97. 0		
Auburn, Ala	First Second.	102 275	96 12	96.5 96	96 96	98.5 98	97 23, 5	97.5 99		
Mobile, Ala	First Second.	140 262	7	94.5	11.5 0	96.5 96.5	75.5 a0	99 97. 5		
Baton Rouge, La	First Second.	· 121 247	90 0,5	93 97.5	91	93.5 65	35 0	96.5 48.5		
San Juan, P. R	First Second.	131	84.5 50	98 96.5						
Wagoner, Ind. T	First Second.	126 238	$93.5 \\ 24.5$	97.5 95	95, 5	97 97. 5	96 34	94.5 97.5		
Durham, N. H	First Second.	147 251	96, 5 0	96 97.5	94.5 96	96 97	93 91	94.5 98		
Ann Arbor, Mich	First Second.		95 97. 5	96 97. 5	99, 5 95	97 96. 5	93 47	97 98		
Average percentage of ger-	first	128	82.19	95, 81	83.79	96.21	81.36	96.64		
mination.	[Second .	251	25.12	96, 25	61	92.36	33.08	90.86		
Average percentage of gain or loss in vitality.	First  Second.	128 251	14.20 74.11	.+0.01 1.20	12, 53 37, 12	+ 0.43 4.80	15.07 65.90	+0.87		

aThis test has not been included in making up the averages inasmuch as the seeds were badly molded when put in test.

The onion seeds which were stored in the envelopes were very seriously affected in many of the places. Those from the basement at Lake City, from all of the conditions at Mobile, and from the dry room and basement at Baton Rouge were entirely killed. The seed from trade conditions at Baton Rouge germinated only 0.5 per cent. In many other cases the samples from the envelopes had become practically worthless. In only two instances was there any loss in vitality where the seeds were stored in bottles, viz, the second tests from the dry rooms and basement at Baton Rouge. These two tests have lowered the average results quite materially. If they were not included the averages would be raised to 96.91 and 97.90 per cent, respectively, instead of 92.36 and 90.86 per cent, as given in the table. The average percentages of germination of the seeds from the envelopes were very low in the second test, and were as follows: Trade conditions, 25.12 per cent; dry rooms, 61 per cent, and basements, 33.8 per cent. This represents a loss in vitality of 74.11, 37.12, and 65.9 per cent, respectively.

Onion seed is relatively short lived, and very easily affected by unfavorable external conditions. For this reason onion seed should be handled with the greatest care if vitality is to be preserved for a maximum period. This may be done successfully by keeping the dry seed in well-corked bottles, or in any good moisture-proof package.

Table XXI.—Percentage of germination of pansy subjected to various conditions of storage in different localities.

[Germination of control sample: First test, 63 per cent; second test, 53 per cent.]

				Perce	ntage of	germin	ation.	
Place of storage.	Order of tests.	Num- ber of days in	Trade conditions.		Dry r	ooms.	Basen	nents.
		storage.	Envel- opes.	Bottles.	Envel- opes.	Bottles.	Envel- opes.	Bottles.
Lake City, Fla	First Second .	129 234	44.5 1.5	63 54	45 22, 5	58.5 47	10.5	62, 5 57, 5
Auburn, Ala	First Second.	102 275	57.5 2	68 20, 5	66.5 28	62 27, 5	60	59. 5 33. 5
Mobile, Ala	First Second .	140 262	3 0	57.5 20.5	2 0	61 25, 5	1	59 2, 5
Baton Rouge, La		121 247	28.5 0	53 34	38 0	44 17	$\frac{4.5}{0}$ .	54 2.5
San Juan, P.R		131	20 6, 5	60.5 58.5				
Wagoner, Ind. T	First Second .	126 238	48.5 7.5	61.5 65	50, 5	62, 5 59, 5	46 8, 5	59 52, 5
Durham, N. H	First	147 251	55, 5 0	66.5. 60.5	49.5 44	63.5 60.5	49 36, 5	63.5 60
Ann Arbor, Mich	First Second.		53.5 46.5	51 45	59.5 52	40 48, 5	50 3, 5	53 60, 5
Average percentage of germination.	{First {Second.	128 251	38.87 8	60, 12 44, 75	44.43 24.41	55, 93 40, 80	31, 57 8, 08	58. 64 38. 43
Average percentage of loss in vitality.	First	128 251	38.3 84.91	4.57 15.60	29.48 53.97	11.23 23.02	49.89 84.76	6, 92 27, 49

Table XXII.—Percentage of germination of phlox drummondii subjected to various conditions of storage in different localities.

[Germination of control sample: First test, 69 per cent; second test, 53.9 per cent.]

				Perce	entage o	f germin	ation.	
Place of storage.	Order of tests.	Num- ber of days in		condi- ons.	Dry rooms.		Baser	nents.
		storage.	Envel- opes.	Bottles.	Envel- opes.	Bottles.	Envel- opes.	Bottles.
Lake City, FlaDo		129 234	41.5 2.5	78 57	62 6	62 25, 5	20.5	77.5 63
Auburn, Ala		102 275	61.5	72.5 56.5	62 13, 5	63 59	65.5 1	67.5 65
Mobile, Ala		140 262	0, 5	55 51.5	0.5	74.5 58.5	0.5	58.5 48.5
Baton Rouge, La		121 247	47.5 0	62.5 58	43.5 0	58, 5 58, 5	$\frac{2}{0}$	70.5 61.5
San Juan, P. R		131	23, 5 11, 5	65 61, 5				
Wagoner, Ind. T		126 238	50.5 5.5	73.5 66	61	70 57	65 9, 5	75 47.5
Durham, N. H		147 251	67 0.5	74 62, 5	62.5 33	45, 5 30, 5	69.5 45.5	71.5 70
Ann Arbor, Mich			67 40	66 54	75, 5 55	69, 5 58, 5	64.5 10.5	72 61
Average percentage of ger-	12	128	44.87	68.31	52.76	63, 28	41.07	70.35
mination.	Second .	251	7, 62	58.37	17.91	49, 64	11.08	59.5
Average percentage of gain	1.5	128	34.97	1	23.54	8, 29	40.49	+ 2.01
or loss in vitality.	Second .	251	85, 86	+8.27	66.78	7. 91	79.45	+10.39

Pansy and phlox have been considered together, since their behavior was almost the same. Both of the controls deteriorated to a considerable degree during the 123 days which elapsed between the time of the first and the second test, pansy losing 15.87 per cent and phlox 21.88 per cent. In both cases the mean loss in vitality of the seeds in the envelopes was very great. The results of the second tests show a loss of 84.91 per cent for pansy, and 85.86 per cent for phlox where stored under trade conditions. In dry rooms there was a mean loss of 53.57 per cent for pansy and 66.78 per cent for phlox, and in basements a loss of 84.76 per cent for the pansy and 79.45 per cent for the phlox. These results are obtained by considering the second test of the control as a standard, the depreciation of the control being disregarded. Some samples were dead and many more were of no economic value. It is especially interesting to note how quickly the seeds died at Mobile, Ala., there being only a few germinable seeds at the end of 140 days.

The behavior of the seeds in the bottles was more or less variable. Some of the pansy seeds showed a higher vitality than the control, but the averages were somewhat lower, the mean loss ranging from 15.60 per cent under trade conditions to 27.49 per cent in basements, while with the phlox the means for trade conditions and for basements were higher than the control by 8.27 and 10.39 per cent, respectively.

Table XXIII.—Percentages of germination of tomato subjected to various conditions of storage in different localities.

[Germination of control sample: First test, 95.5 per cent; second test, 97.5 per cent.]

				Perce	ntage of	germin	ntion.	
Place of storage.	Order of tests.	Num- ber of days in	Trade conditions.		Dry r	ooms.	Baser	nents.
		storage.	Envel- opes.	Bottles.	Envel- opes.	Bottles.	Envel- opes.	Bottles.
Lake City, Fla	First Second.	129 234	94 94	94 98	94 94	95. 5 97. 5	88.5 77	94 97.5
Auburn, Ala	First Second.	102 275	95 94	94.5 98.5	93.5 97	97.5 94.5	96 98	94.5 96.5
Mobile, Ala	First Second.	140 262	90 79.5	94.5 97.5	91.5 87	96.5 95.5	64.5 19.5	93.5 98
Baton Rouge, La	First Second.	121 247	91.5 96	95 96. 5	91 93	95 98	83.5 39.5	95 96
San Juan, P. R	First Second .	131	94 96, 5	94.5 94.5				
Wagoner, Ind. T	First		96.5 94	97 98	98	96.5 97.5	98.5 98.5	96 93.5
Durham, N. H	First:		94.5 87	95 98	97 97	94 99	97.5 97.5	96.5 97
Ann Arbor, Mich	First Second.		89 98. 5	94 98	93 98	91.5 97.5	89 95	92.5 98
Average percentage of ger-	First	128	93.06	94.81	84	95. 21	88.21	94, 57
mination.	[Second .	251	92.44	97.31	94.33	97.07	84, 25	97, 21
Average percentage of loss	First	128	2.56	0.72	1.57	0.30	7.64	0.98
in vitality.	Second.	251	5. 20	0.20	3, 29	0.44	13.63	0.30

The tomato seed, as shown in Tables V and XXV, was the most resistant to the unfavorable conditions of storage. The seed in the bottles was not injured at any of the places. The lowest germination was 91.5 per cent from the seed kept in a dry room at Ann Arbor, Mich. The seed in the envelopes gave a much wider variation, falling quite low in some of the samples which were stored in the basements. The average losses in vitality for the entire series of the second set of seeds which were kept in envelopes were as follows: Trade conditions, 5.20 per cent; dry rooms, 3.29 per cent; basements, 13.63 per cent. The average percentage of germination of the seed which was kept in the bottles differed from the control less than one-half of 1 per cent.

Table XXIV.—Percentage of germination of watermelon subjected to various conditions of storage in different localities.

Germination of control	sample: First tes	t 95.5 ner cent	; second test, 99 per cent.]

				Perce	ntage of	germin	ation.	
Place of storage.	Order of tests.	Num- ber of days in			Dry r	ooms.	Baser	nents.
		storage.	Envel- opes.	Bottles.	Envel- opes.	Bottles.	Envel- opes.	Bottles.
Lake City, Fla	First Second.	129 234	98 92	98 96, 2	96 86	98 98	98 70	100 91
Auburn, Ala	First Second.	102 275	94 86	94 100	96 98	98 98	99 94	100 96
Mobile AlaDo	First Second.	140 262	98 64	98 96	98 68	100 96	80	100 100
Baton Rouge, La	First Second.	121 247	100 92	98 98	96 86	100 100	98 20	98 100
San Juan, P. R.	First	131	96 88	100 100		l		l
Wagoner, Ind. T	First Second.	126 238	98 94	98 98	98	100 96	96 88	98 98
Durham, N. H	First Second.	147 251	98 82	98 96	100 98	98 92	98 94.1	96 98
Ann Arbor, Mich	First Second .		100 96	100 100	94 96	94 92	98 100	96 96
Average percentage of ger-	[First	128	97, 75	98	96,86	98.29	95.29	98, 29
mination.	Second .	251	86.75	98.02	88.67	96	77.70	97.43
Average pecentage of loss	{First	128	0.56	0.31	1.47	0.01	3.06	0.01
in vitality.	(Second.	251	12.37	0.99	10.44	3.03	21.52	1.59

What has been said of the tomato seed is practically true for the watermelon, save that there was a greater loss in vitality in the latter, when seeds were kept in envelopes. The average percentage of germination of the second tests was 86.75 per cent for trade conditions; 88.67 per cent for dry rooms; and 77.7 per cent for basements, or a loss in vitality of 12.37, 10.44 and 21.52 per cent, respectively, as compared with the vitality of the control sample, which germinated 99 per cent.

An examination of the foregoing set of tables will show that in most cases the deterioration was comparatively slight during the first 128 days. Yet even during this short period the losses in vitality were very marked in some of the more critical localities, particularly at Mobile. However, the greatest loss, as shown by the germination tests, was during the 123 days immediately following.

While seeds, like other living things, are capable of withstanding quite unfavorable conditions for a considerable time without showing any appreciable deterioration in vitality, still the forces destroying vitality are at work. When the turning point is once reached and can be detected by germination tests, the decline is more noticeable and death soon follows.

The preceding tables show that the loss in vitality was very different in the different places. The conditions at Mobile, Ala., proved to be the most injurious, while those at Ann Arbor, Mich., were the most conducive to longevity. These results, however, are given in another part of this paper dealing with the effect of climate on the vitality of seeds. The results are tabulated on pages 18 and 23 and represented diagrammatically on page 24, so that any further discussion at this time is unnecessary.

Likewise each table has been summarized, giving the average percentages of germination and the average percentages of the loss in vitality of each sample of seed for both the first and second tests. These averages include those of the three conditions of storage—trade conditions, dry rooms, and basements—in both envelopes and bottles.

Naturally, the results of the second tests are of the greater importance, and, in order that the results may be readily compared and more critically examined, they have been collected and tabulated herewith:

Table XXV.—Average percentage of germination and average percentages of loss in vitality of the different kinds of seeds when kept under different conditions.

0 0		**											
	ol	Tra	ide co	nditio	ns.		Dry re	ooms.			Baser	nents.	
	control	Envel	opes.	Bott	les.	Envel	opes.	Bott	les.	Envel	opes.	Bot	tles.
Kind of seed.	Germination of c	Germination.	Loss in vitality.										
Tomato	97.5	92, 44	5, 20	97.31	0.20	94, 33	3, 29	97.07	0.44	84.25	13.63	97.21	0.30
Sweet corn, "A"	92.4	83	10.11	96.75	+4.71	83, 33	9.81	94.86	+2.66	73.08	4)1)	98	+ 6.06
Peas	95.7	84.74	11.45	95, 25	. 47	80,45	15, 94	95.14					
Watermelon	99	86.75	12.37	98.02	. 99	88.67	10.44	96	3, 03	77.70	21.52	97.43	1.59
Lettuce	92.3	77.75	15.76	91.12	1.29	78, 33	15, 14	90.93	1.49	65.58	28.95	90.78	1.65
Radish	78.8	60.94	22.67	73.56	6, 65	64.33	18.37	72.71	7.73	59	25.13	74.07	6
Sweet corn, "B"	88.5	65.41	26.09	59.70	32, 55	66.33	25, 06	48	45.76	60.41	31.74	68.40	22.71
Bean	98.7	69,50	29.59	97	1.72	69.33	29.76	97.36	1.36	55.66	43.61	98, 86	+.10
Cabbage	92.4	52.15	43.56	90.56	1.94	61.50	33, 44	89, 93	2.67	53,33	42, 29	92, 21	. 22
Carrot	82	37, 31	54.50	80.87	1.38	53, 83	34.35	74.71	8.89	37, 75	53, 96	75.21	9.50
Onion	97	25.12	74.11	96, 25	1.20	61	37.12	92, 36	4.80	38.08	65, 90	90.86	6, 33
Pansy	53	8	84.91	44.75	15,60	24, 41	53.97	40,80	23.02	8.08	84.76	38.43	27.49
Phlox	53.9	7.62	85, 86	58.37	+8.27	17.91	66.78	49.61	7.91	11.08	79.45	59, 50	+10.39
Average loss in vitality .			36, 63		3, 92		21.19		8, 08		42.28		4, 51

In comparing the average results shown in Table XXV, it will be seen that different seeds behave very differently under practically identical conditions. The list of seeds has been arranged according to their loss of vitality as represented by those kept in envelopes under trade conditions, as shown in the fourth column. The tomato seed gave a loss in vitality of 5.20 per cent, being the most resistant to the unfavorable climatic conditions. Phlox, on the other hand, germinated only 7.62 per cent, representing a loss in vitality of 85.86 per cent.

Likewise the same seeds behave very differently under slightly different conditions, as will be seen by comparing the percentages of deterioration in the case of seeds kept in envelopes under trade conditions, in dry rooms, and in basements. In dry rooms the order, except the peas, is the same as for trade conditions. The loss of vitality in the seeds stored in the dry rooms was uniformly less than for those stored under trade conditions, excepting for the peas and beans; but in the series from the basements there was great irregularity. The loss in vitality for the most part was uniformly greater than under trade conditions or in dry rooms save in the last five—eabbage, carrot, onion, pansy, and phlox—where the loss was less in the case of those kept in the basements. This indicates that these five species of seed are less susceptible to the evil effects of a moist atmosphere when the temperature is relatively low.

The relative value of these three conditions for storing seeds in paper packets is best obtained by a comparison of the general averages. The average losses in vitality for the thirteen different samples of seed which were kept at the eight different stations were as follows: Trade conditions, 36.63 per cent; dry rooms, 21.19 per cent; basements, 42.28 per cent. From these results it is quite clear that seeds put up in paper packages will retain their vitality much better if kept in dry, artificially heated rooms than if they are subjected to trade conditions or stored in basements.

But another comparison needs yet to be made, and is the most important of the series, i. e., the vitality of seeds when kept in closely corked bottles. In the majority of cases there was but little deviation from the control samples, and many of the samples germinated even better where the seeds were kept in bottles. The "A" sweet corn offers the best illustration of the increased germination. At the same time the "B" sample of sweet corn was very much injured. Here are two samples of the same variety of corn behaving very differently when kept in bottles. This difference in vitality is directly attributed to the greater quantity of water in sample "B," showing the necessity of thoroughly drying seeds if they are to be put up in closed vessels. A comparison of the general averages of the bottle samples and of those kept in envelopes indicates that the former is far superior to the latter as a method for preserving the vitality of seeds. Under trade conditions the loss in vitality was 36.63 per cent in envelopes and

3.93 per cent in bottles; in dry rooms, 21.19 per cent in envelopes and 8.08 per cent in bottles; in basements, 42.28 per cent in envelopes and 4.51 per cent in bottles.

The necessary precautions to be taken, if seeds are to be stored in bottles, are (1) a well-dried sample, preferably artificially dried seed, and (2) a cool place for storing, at least a place in which the temperature will not be higher than the temperature at which the seeds were originally dried.

If the above precautions are taken at least two beneficial results will follow: First, protection against moisture, which is of considerable importance, as many seeds are soon destroyed in that way when kept in paper packages. Secondly, vitality will be preserved for a longer period and consequently there will be a more vigorous germination, a better growth of seedlings, and a greater uniformity in the resulting crop.

Having thus shown that seeds retain their vitality in warm, moist climates much better when kept in bottles than when kept in paper packages, the necessity of finding a more suitable method for sending small quantities of seed to such places at once presents itself.

# EXPERIMENTS IN KEEPING AND SHIPPING SEEDS IN SPECIAL PACKAGES.

At present the greatest disadvantages in sending out seeds in bottles are the inconvenience and expense involved by this method of putting up seeds. The increased cost of bottles, as compared with the paper packets now so universally employed, the additional labor and expense necessary to put up the seeds, the greater cost in handling and packing the bottles to insure against losses by breakage, and the increased cost of transportation, are all matters of vital importance. Seedsmen claim that the existing conditions of the trade will not admit of their raising the price of seeds sufficiently high to justify the increased expense of glass containers. Although to the seedsmen the preservation or the prolongation of vitality is an important factor, yet the demand is for an inexpensive and at the same time a neat and serviceable package.

Accordingly, duplicate samples of the following-named seeds were put up in special packages, one set being sent to Mobile, Ala., and the other kept at Ann Arbor, Mich. The seeds used for these experiments were beans, peas, cabbage, lettuce, onion, pansy, and phlox.<sup>a</sup>

a The lettuce, onion, pansy, and phlox were from the same bulk samples of seeds as those used in the earlier experiments; but the beans, peas, and cabbage used for these tests were from samples received at the laboratory on February 4, 1901. However, the latter three were from the same general stock of seed, differing from those used in experiments already given only in that they were stored during the interval in the warehouse of D. M. Ferry & Co., Detroit, Mich., instead of in the botanical laboratory at the university.

All of these samples were first dried for ten days in an incubator maintained at a temperature of from 30° to 32° C. The amount of moisture in the samples before and after drying, as well as the moisture expelled during the drying process, was as follows:

Moisture test of seeds in special packages.

Kind of seed.	Moisture in air-dried samples.	Moisture remaining.	Moisture liberated.
	Per cent.	Per cent.	Per cent.
Beans	10.32	4.90	5.42
Peas	9.70	6, 00	3.70
Cabbage	4.89	3.47	1.42
Lettuce	5.33	3,80	1.53
Onion	6,48	4. 17	2.01
Pansy	4.82	3.13	1.69
Phlox	5, 82	4.30	1.52

These well-dried seeds were then put up in seven different kinds of packages:

- (1) Double coin envelopes, of much the same quality as those in which seeds are commonly sold.
  - (2) Bottles of 120 cc. capacity, closed with firm cork stoppers.
  - (3) Bottles of 120 cc. capacity, corked and sealed with paraffin.
- (4) Tin cans having closely fitting lids, the whole being then carefully dipped in paraffin.
- (5) Double coin envelopes, as for No. 1, the packets being then dipped in melted paraffin.
- (6) Double coin envelopes, the inner one paraffined, the outer envelope being used simply to protect the paraffin and to facilitate ease of handling.
- (7) Double coin envelopes, with both the inner and the outer coated with paraffin.

On February 15, 1901, one of each of the above preparations was sent to Mobile, Ala., and stored in a cellar approximately 400 feet back from the bay. After the lapse of 108 days, i. e., on June 3, these samples were received in return, at which time germination tests were made.

The other complete set, retained in the botanical laboratory at Ann Arbor, was subjected to a very moist atmosphere. The samples were kept in a damp chamber made by taking two battery jars of different sizes, the smaller containing the seeds being placed within the larger, which was lined with filter paper and then partially filled with water. The whole was covered with a glass plate, and the atmosphere within was always on the verge of saturation.

A third and an extreme set of conditions was established by keeping some of the paraffined packages immersed in water for twenty-seven

days. At the end of that time (March 14) the seeds were tested for germination, as were also those from the unprotected envelopes in the moist chamber. The seeds that were kept under water in the paraffined packages germinated readily and normally, showing no deterioration in vitality; but the seeds from the packages not paraffined. which were kept in the moist chamber, had been injured to an appreciable extent, there being a marked retardation in the germination of all of the species of seed. The cabbage at the end of thirty-six hours had germinated only 11 per cent, as compared with 57.5 per cent for seed from the immersed paraffined package. The relative merits of the two conditions as affecting onion seed may be expressed by a germination of 13.5 per cent and 39 per cent, respectively, after sixtyone and one-half hours. Not only was there a marked retardation, but likewise a reduction in the final percentage of germination, with the single exception of the cabbage. These results can be more carefully studied in Table XXVI.

Germination tests were made of all of the other samples on June 3, 1901, the date when the seeds were returned from Mobile. At this time the seeds in the unprotected envelopes in the moist chamber were so badly molded that no germination tests were made. The samples from Mobile, which were directly comparable with the above, except that they had been stored in a basement, were greatly injured. The beans had deteriorated to 88 per cent, the onion to 27 per cent, the pansy to 8 per cent, while the phlox was dead. However, seed of the other species—cabbage, lettuce, and peas—gave final percentages of germination varying but little from the control, but the slowing down in the rapidity of germination was sufficiently marked to show a corresponding loss in vitality.

With the samples which were put up in bottles, tin cans, and paraffined packages the results were quite different from those given above. In no case was there any marked deviation beyond that which might be justly attributed to ordinary variation, except in the phlox from a tin can which had been stored in the moist chamber in the laboratory. This sample of phlox germinated only 3.5 per cent. Unfortunately, both the pansy and the phlox seeds used for these experiments were not very satisfactory. These samples were at this time nearly two years old and consequently of a low vitality. The tabulated results of the foregoing experiment follow.

Table XXVI.—Vitality of seeds preserved in different kinds of packages.

	Dura-		Pe	ercent	age of	germi	nation	١.	
Treatment of samples.	cion of experi- ment.	Beans.	Cab- bage.	Let- tuce.	On- ions.	Peas.	Pan- sy,	Phlox.	Aver- ages.
	Days.								
Control		94.0	90.2	89, 5	97, 5	90.0	37.7	42.5	77.34
Ann Arbor, Mich., moist chamber:									
Envelopes	27	80.0	91.0	76.5	90.0	88.0	25.0	0.0	64.35
Bottle, corked	108	98.0	91.5	91.0	93.5	94.0	36.0	31.0	76.43
Bottle, paraffined	108	97.5	93, 5	90.5	95.5	90.0	39.5	39.0	77.9
Tin can, paraffined	108	96.0	87.0	90.0	93.0	90.0	35.0	3, 5	70.6
Two envelopes, outer paraffined	108	98.0	91.5	91.5	97.0	92.0	33.5	27.5	75.8
Two envelopes, inner paraffined	108	98.0	94.0	89.0	93. 0	88.0	24.0	47.0	76.1
Two envelopes, both paraffined	108	96.0	90.5	86.5	95.5	92.0	23.0	38.5	74.5
Two envelopes, both paraffined and									
immersed in water	27	100, 0	88, 5	88.5	94.5	90.0	34.5	30.5	75, 2
Mobile, Ala., basement:									
Envelopes	108	88.0	86.0	88.0	27.0	96.0	8.0	0.0	56.1
Bottle, corked	108	98. 0	91.0	90.5	95.5	84.0	34.5	32.5	75.1
Bottle, paraffined	108	98.0	90.5	92.5	95.5	92.0	34.5	44.5	78. 2
Tin can, paraffined	108	96. 0	88.0	95.0	96.0	88.0	26.0	23.0	73.1
Two envelopes, outer paraflined	108	94.0	90.5	89.0	95.5	92.0	29.5	34.0	74. 7
Two envelopes, inner paraffined	108	96, 0	92.0	88. 0	90.0	98.0	33, 0	38, 0	76.4
Two envelopes, both paraffined	108	100.0	92.0		88.5		25, 5	33.5	74.1

Subsequent experiments were made, using envelopes of different qualities, as well as varying the treatment of the packages. Samples of cabbage, lettuce, and onion seed were put up as follows:

- (a) The regular seedsmen's envelope, made of a heavy grade of manila paper.
  - (b) Envelopes made of a medium quality of waterproof paper.
  - (c) Envelopes made of a thin parchment paper.
- (d) Envelopes made of the same quality of parchment paper as for the preceding series, but paraffined previous to being filled with seed. The packages were then sealed by redipping the open ends.
- (c) Envelopes of parchment paper, as for the two preceding series, except that the envelopes were first filled with seed, sealed, and then the entire package was dipped in paraffin at a temperature of from 55 to 60 (d.

Samples of all of these packages were then stored under trade conditions and in dry rooms in Ann Arbor, Baton Rouge, and Mobile. The exact conditions of storage in the different places were the same as described on pages 49 and 50.

The samples were put up on May 20, 1901. The period of storage ended on November 26, having continued 190 days. Unfortunately, no special precautions were taken to dry the seeds. They were simply air-dried samples; hence they contained a quantity of moisture sufficiently large to give rise to an increased relative humidity of the confined air in the paraffined packages. This increased humidity was

accompanied by a greater activity within the cells, and consequently by a greater deterioration of vital force. For this reason the results are not as definite as the conditions warrant. Nevertheless, some important facts were brought out by the experiments which justify their being discussed and tabulated (in part) at this time.

Table XXVII.—Vitality of seed preserved in paraffined packages.

	Trade cond	itions, seeds	put up in—	Dry roo	m, seeds put	up in—
Kind of seed.	Paraffined envelopes.	Parchment envelopes, then dip- ped in par- affin, at 50° to 60° C.	Seedsmen's packages.	envelopes.		Seedsmen's packages.
Cabbage:	Per cent.	Per cent.	Per cent.	Per cent.	Per cent.	Per cent.
Ann Arbor, Mich	91	90	86.5	90.5	85, 5	86.5
Mobile, Ala	30, 5	57.5	8,5	38	50.5	5
Baton Rouge, La	70	63	22.5	73.5	79.5	35.5
Lettuce:						
Ann Arbor, Mich	89.5	89.5	96.5	91.5	90	93
Mobile, Ala	80	75	64	. 78	78.5	61.5
Baton Rouge, La	81.5	77.5	74	82	73.5	72.5
Onion:						
Ann Arbor, Mich	91	90	93	91, 5	89	89
Mobile, Ala	0	4	0	0	4.5	0
Baton Rouge, La	1	20	0	5	40	0
Average	59.39	62.94	49, 44	61.11	65, 66	49. 22

In the first place, the injury resulting from the effect of the climatic influences is quite well marked in the above table. The conditions at Mobile and Baton Rouge were much more detrimental to the life of the seeds than were the conditions at Ann Arbor. Secondly, the differences in the preservation of vitality of those seeds stored under trade conditions and of those kept in dry rooms were much less marked than they were in earlier experiments. This is probably accounted for by the marked difference in the two seasons. The summer of 1900 was extremely wet in the South, especially at Mobile, while the summer of 1901 was exceptionally dry. Concerning the conditions Zimmer Brothers wrote on November 26, 1901, as follows:

We do not think you will find much difference in the two packages. The season this year has been very dry, with no rain since the big August storm; in fact, we do not remember such a dry season in thirty years.

Although the season was exceptionally dry at Baton Rouge and Mobile, the loss in vitality was very great in comparison with the loss at Ann Arbor, demonstrating very clearly that climatic influences play a very important part in the storage of seeds.

This table shows the relative resisting powers of lettuce, cabbage, and onion seed, the lettuce being most resistant and the onion least resistant, as shown in a preceding table. However, the chief purpose

of this series of experiments was to demonstrate the relative value of different packages as a means of putting up seeds.

In Table XXVII it will be observed that the results obtained from the waterproof and parchment paper envelopes have been omitted. These omissions have been made because the results were practically identical with those of the ordinary seedsmen's packets; but the comparisons to be made between the ordinary paper packets and the paraffined packages are worthy of consideration. The envelopes that were paraffined after being filled with seed gave the best results. This difference, however, was due not to the special treatment but to the higher melting point of the paraffin. The average percentages of germination of the three samples of seed kept under trade conditions in the three localities were 59.39 per cent for the envelopes previously paraffined, 62.94 per cent for the envelopes dipped in paraffin after being filled with seed, and 49.44 per cent for the seedsmen's envelopes. In dry rooms the results were 61.11, 65.66, and 49.22 per cent, respectively. These averages were somewhat higher than the true conditions of Baton Rouge and Mobile warrant, as the results of the germination tests from all of the packages retained at Ann Arbor showed but little variation. Taking the three samples of seed which were stored under trade conditions in Mobile, the average percentage of germination was 24.2 for the seed from the nonparaffined package and 45.5 per cent for the seed from the paraffined package, showing a loss in vitality of 77.3 and 49.5 per cent, respectively, considering the germination of the Ann Arbor sample as a standard. At Baton Rouge the results were slightly better; the average percentages of germination were 32.2 for the seeds from the nonparaffined and 53.5 per cent for the seeds from the paraffined packages, representing a loss in vitality of 65 and 40.5 per cent, respectively. While in either case the loss was very great, still the advantages of the paraffined packages are worthy of consideration for the reason that a prolongation of life for only a few weeks is frequently of the greatest importance, particularly in districts where much fall planting is done.

In this connection may be given the results of some other tests, which really were a part of this same experiment, but included only onion seed. This seed was put up in seedsmen's envelopes and in paraffined envelopes like those previously described. In addition, seed was also put up in small bottles, which were corked. These packages were kept in a small box within a suit case carried on two trips across the Atlantic and on a tour through Central Europe, thus subjecting them to very variable conditions. Germination tests gave the following results: Seed from the ordinary packages, 77 per cent; paraffined envelopes, 90 per cent; bottles, 91 per cent.

To test more thoroughly the keeping qualities of seeds in paraffined packages and in bottles, another series of experiments was begun on December 20, 1901. For these tests only cabbage and onion seeds

were used, but each with three different degrees of moisture: (1) Seed from the original packages, i. e., air-dried samples, the cabbage having a water content of 5.80 per cent, and the onion 6.48 per cent. (2) Air-dried samples were exposed in a moist atmosphere under a bell jar for two days, during which time the cabbage absorbed 1.83 per cent of water and the onion 2.41 per cent, thus raising the water content to 7.63 and 8.89 per cent, respectively. (3) Air-dried seeds which were dried in an incubator for eight days at a temperature varying from 27 °C. to 39°C. During this interval 2.05 per cent of water was expelled from the cabbage and 3.11 per cent from the onion seed, leaving a water content of only 3.75 per cent in the former and 3.37 per cent in the latter.

Each of the samples, treated as just described, was put up in three different kinds of packages: (1) Seedsmen's regular seed envelopes. (2) Similar envelopes which were paraffined, after being filled with seed, at a temperature of from 70° to 75° C. The melting point of the paraffin was 53° C. (3) In bottles which were closed with firm cork stoppers.

One of each of the above packages was then stored at Mobile under trade conditions and in a basement; likewise at Ann Arbor in the herbarium room of the botanical laboratory, in a greenhouse, and in an incubator maintained at  $40^{\circ}$  C. The duration of this experiment was 131 days, from December 20, 1901, to April 30, 1902. The results of the germination tests are given in Table XXVIII. Two percentages have been given for the control sample, one for Ann Arbor and the other for Mobile. This was necessary since the two series were tested at different times and comparisons can not be made interchangeably between the two.

Table XXVIII.—Vitality of cabbage and onion seed as preserved in various kinds of packages and subjected to different conditions of storage.

[Germination of control samples—Ann	Arbor: Cabbage	81.7 per	cent; onion, 7	4 per cent.	Mobile:
Cabbage, 8	8 per cent; onion	, 84.5 per o	cent.]		

				Percentage of germination.									
Kind of seed and	Special treat-	Percent- age of		Ann Ar	Mobile, Ala.								
package.	ment of package.	water content of seed.	Botan- ical labo- ratory.	Trade conditions.	Green- house.	Incuba- tor at 40° C.	Trade condi- tions.	Base- ment.					
Cabbage:													
Envelope	None	5, 80	81.0	81.0	68.0	72.5	60.0	10.0					
Do	Paraffin	5, 80	, 80.0	79.0	85.5	62, 0	87.5	52.5					
Bottle	Corked	5, 80	79.5	85, 0	85.0	68.5	84.0	84.0					
Envelope	None	7.63	85.5	80.5	65, 5	74.5	64.5	15.5					
Do	Paraffin	7.63	80.5	82.0	83.5	69.5	86.5	46.5					
Bottle	Corked	7.63	80.5	85.0	86.5	48.0	82.0	91.5					
Envelope	None	3.75	76.0	85.5	67.0	73.0	64.0	9.0					
Do	Paraffin	3.75	86.0	84.0	76.0	71.0	82.5	78.0					
Bottle	Corked	3.75	83.0	84.0	74.0	64.5	82.5	85.0					

Table XXVIII.—Vitality of cabbage and onion seed as preserved in various kinds of packages and subjected to different kinds of storage—Continued.

Account of the second		Percent- age of	Percentage of germinatien.								
Kind of seed and	Special treat-			Ann Ar	h.	Mobile, Ala.					
package.	ment of package.	water content of seed.	Botan- ical labo- ratory.	Trade conditions.	Green- house.	Incuba- tor at 40° C.	Trade conditions,	Base- ment.			
Onion:											
Envelope	None	6, 48	78.5	69.5	3.5	47.0	19.5	10.0			
Do	Paraffin	6,48	76.5	66,5	67.0	4.5	83.0	27.0			
Bottle	Corked	6.48	73.5	71.5	60, 0	64.0	86.0	82.5			
Envelope	None	8, 89	74.5	60.0	11.5	28.0	21.0	2.5			
Do	Paraffin	8, 89	74.5	66, 0	56, 0	9, 0	74.5 [	21.0			
Bottle	Corked	8.89	78.0	68.0	67.5	3, 0	77.5	78, 5			
Envelope	None	3.37	61.5	63.5	8, 5	? 6.0	17.0	6.0			
Do	Paraffin	3,37	75.5	72.5	. 58, 0	? 9.0	77.0	60.5			
Bottle	Corked	3.37	76, 5	71.0	77.0	59.5	81.5	81.5			
								_			

Many of the points brought out by this table are very similar to those of the preceding one, yet the differences are sufficiently marked to justify its being given in this connection. The seeds stored in the botanical laboratory and those subjected to trade conditions at Ann Arbor have germinated practically the same, the cabbage slightly favoring trade conditions and the onion being better preserved in the laboratory. But a comparison of the trade conditions at Ann Arbor and Mobile in the unprotected packages shows the same wide variation that has been already pointed out.

The advantage of drying is not very clearly brought out in this table; in many cases there seems to have been a slight injury as a result of the high temperature at which the drying was done. Unavoidably the temperature at that time reached 39° C., which, as has already been stated, is slightly above the maximum to which seeds can be subjected for any considerable time without injury. The injury due to heat is very evident in the samples stored in the incubator maintained at 40° C., this injury being more apparent with the increased moisture, especially in the paraffined package and in the bottle. However, on the whole the percentages of germination are higher for the dried seed than for the seed which had absorbed an additional quantity of moisture; and, indeed, the comparison should properly be made with these two, for seeds as they are usually stored contain even higher percentages of moisture than either the cabbage or lettuce after they had absorbed the additional amount of water.

But the chief purpose of the present experiments was to determine the relative advantages of envelopes, paraffined packages, and bottles as methods of putting up seed in order that vitality might be preserved for a longer time. This comparison is best made by considering the vitality of the seed stored in the greenhouse at Ann Arbor and under trade conditions at Mobile. It will be readily seen that the vitality of the seed from the unprotected packages was greatly reduced, while those from the paraffined envelopes and from the bottles germinated nearly as well as the controls. These differences are better represented diagrammatically, as follows:

Diagram representing the percentages of germination of cabbage seed when treated as described.

Kind of package.	Special treatment of package.	Percentage of water content of seeds.	Ann Arbor, Mich., green- house.	Mobile, Ala., trade conditions.
Envelope		5, 80	73.3	60
Do	Paraffined	5, 80	92.1	87.5
Bottle	Corked	5, 80	91, 5	81
Envelope		7, 63	70.5	64.5
Do	Paraffined	7.63	89. 9	86.5
Bottle	Corked	7.63	93, 1	82
Envelope		3.75	72.1	64
Do	Paraffined	3.75	81.8	82.5
Bottle	Corked	3, 75	79.7	82.5
Control sample .	Original package.	5, 80	88	88

Diagram representing the percentages of germination of onion seed when treated as described.

Kind of package.	Special treat- ment of package.	Percentage of water content of seeds.	Ann Arbor, Mich., green- house.	Mobile, Ala., trade conditions.
Envelope		6,48	4	19.5
Do	Paraffined	6.48	76.6	83
Bottle	Corked	6,48	68, 6	86
Envelope		8,89	13.2	П
Do	Paraffined	8.89	64	74.5
Bottle	Corked	8.89	77.3	77.5
Envelope		3, 37	9.7	17
Do	Paraffined	3.37	66.3	77
Bottle	Corked	3.37	88	84.5
Control sample	Original package.	6.48	84,5	84.5

The percentages for Ann Arbor shown in the graphic representations are not the same as those given in the foregoing table. In the diagram they are directly comparable with the results from the Mobile series,

all being based on the vitality of the controls, as shown by the tests made at that time, the standard being 88 per cent for the cabbage and 84.5 per cent for the onion.

A discussion here hardly seems necessary, as there can be no doubt that seeds retain their vitality much better in moist climates if protected from the action of the atmosphere. This may be accomplished by dipping the packages in paraffin or by putting the seed in bottles. Disregarding the expense, bottles surpass paraffined envelopes as a means for the preservation of vitality, and also in the ease with which the seed can be put up. The results are more certain if care is exercised in selecting good corks.

#### RESPIRATION OF SEEDS.

From a practical point of view it has been conclusively shown that moisture is the controlling factor in seed life. Seeds stored in a humid atmosphere soon lose their vitality, but if carefully dried and protected from moisture life is greatly prolonged.

The question at once presents itself: In what way does the presence of increased quantities of moisture cause a premature death of the seed, or why is vitality prolonged if the water content of the seed be reduced?

In a measure, the answer to this question is respiration. Seeds as we commonly know them absorb oxygen and give off carbon dioxid; that is, respire.<sup>a</sup> During their respiratory activities the energy stored within the seed is readily evolved, the vital processes are destroyed, and life becomes extinct. The intensity with which respiration takes place is largely dependent upon the humidity of the surrounding atmosphere, which ultimately resolves itself into the amount of water in the seed. The respiratory activity is directly proportional to the quantity of moisture absorbed by the seed up to a certain point, attaining its maximum during the process of germination. It has been found that a decrease in the water content results in a corresponding diminution in the intensity of respiration and consequently in a prolongation of the life of the seed as such.

Bonnier and Mangin<sup>b</sup> were the first to show that respiration in living plants increases with an increase in the humidity in the surrounding air. As this is true for growing plants, it is even more marked in stored seeds. Maquenne<sup>c</sup> suggested that a reduction in moisture is accompanied by a reduction in respiration, but at that time no experiments had been made to show that such was actually the case.

a Kolkwitz (Ber. d. deutsch. Bot. Ges., 19: 285-287, 1901) reports respiration in recently ground seeds.

<sup>&</sup>lt;sup>b</sup> Ann. sc. nat. bot., ser. 7, 2: 365-380, 1885.

c Ann. Agron., 26: 321-332, 1900.

In 1832, Aug. Pyr. De Candolle wrote in the second volume of his Physiologie Végétale that the vitality of seeds would be prolonged if they were buried sufficiently deep in the soil to protect them from oxygen (or air) and moisture. Unfortunately, De Candolle did not discover the true cause of this prolonged life, for nowhere did he make any reference to respiration. Nevertheless his general conclusions were properly drawn. De Candolle also stated that light accelerates evaporation in seeds and thus causes a premature death. Here, however, his results were wrongfully interpreted. These conclusions are applicable only in case of seeds that die if allowed to become dry. The real effect of light is to cause a slightly accelerated respiration and consequently a greater deterioration in vitality. Jodin a states that light accelerates respiration to a marked degree. His experiments were with peas which contained 10 to 12 per cent of moisture. Two samples of peas were placed, each under a bell jar, over mercury. One sample was kept in the light and the other in the dark. At the end of 4 years 6 months and 14 days an analysis of the confined air from the sample kept in the light gave the following results:

Peas, 3.452 grams, in air, in light:	Per	cent.
Oxygen		19. 1
Nitrogen		78.6
Carbon dioxid		1.2

At the end of 4 years 7 months and 14 days an analysis of a sample of air taken from the other chamber was as follows:

Peas, 3.580 grams, in air, in dark:	Per cent.
Oxygen	20.8
Nitrogen	79.1
Carbon dioxid.	1

The 3.452 grams of peas that were subjected to the influence of the action of light had absorbed, in the given time, 2.4 cc. of oxygen and produced 1.8 cc. of carbon dioxid. The seed kept in the dark showed but little signs of respiratory activity. Germination tests of the former showed the peas to be dead, while five peas from the sample kept in the dark germinated perfectly.

While there is no question that light exerts some influence on respiration, still the above results do not furnish sufficient data to establish the fact that respiration practically ceases in the absence of light. In fact, experiments have shown that respiration is also quite marked in case of seeds stored in the dark, and the difference is very slight if the same temperature be maintained.

Van Tieghem and Bonnier, in their "Recherches sur la vie latente des graines," demonstrated that 7.976 grams of peas, sealed, in air,

a Ann. Agron., 23: 433-471, 1897.

<sup>&</sup>lt;sup>b</sup> Bul. Soc. bot. France, 29: 25-29, 1882.

in a tube, respired quite freely. After the lapse of two years an analysis of the confined air gave the following results:

·	Per	cent.
Oxygen	1	4.44
Nitrogen	8	31.74
Carbon dioxid		3.82

These same seeds germinated 45 per cent and had increased  $_{790}$  of their original weight.

In the experiments of the writer it was found that 40.1150 grams of air-dried beans liberated 7.7 cc. of carbon dioxid in 370 days. The concentration of the carbon dioxid in the flask at the time the gas was drawn for analysis was 1.54 per cent. This sample of seed germinated 97 per cent, and there was only a very slight retardation in germination, which indicated that the vitality had not been materially reduced. During this time there was a slight decrease in the weight of the seed—0.19 per cent. At the same time two check bottles were set up, one containing 40.1184 grams of beans known to be dead, and the other bottle containing nothing except air. Analyses of the air from these two bottles gave the same results as samples of air drawn from the laboratory. These preparations were kept in subdued light throughout the experiment.

That respiration may take place in the dark, that it is very intense if much moisture be present, and that intensive respiration is accompanied by a rapid loss in vitality is shown by the following experiments. On April 3, 1900, samples of beans, cabbage, carrot, lettuce, and onion were sealed, each in bottles of 250 cc. capacity, and were stored in a dark room which was maintained at a temperature of from 20 to 25 °C. These samples were first carefully weighed and then placed in a damp chamber for 175 hours, so that an additional quantity of moisture could be absorbed.

Control samples of air-dried seeds were also kept in sealed bottles and subjected to the same subsequent treatment. After the lapse of one year analyses of the confined gases and germination tests of the seeds were made, the results of which are given with the general details.

Beans,—Of beans, 24.9994 grams absorbed 4.70 per cent of water while in the damp chamber. The respiration during the year was equivalent to 2.5 cc. of carbon dioxid. The loss in weight was only 0.05 per cent, but the vitality had fallen from 100 to 86 per cent, as shown by the control.

Cabbage.—Of cabbage seed, 10 grams, with an additional 9.79 per cent of water, were used for this test. During the year this sample of cabbage seed had given off 24 cc. of carbon dioxid, an equivalent of 2.4 cc. of carbon dioxid per gram of seed per year. The control sample germinated 89 per cent, but this seed was dead.

Carrot.—Of carrot seed, 10 grams were allowed to absorb during 175 hours an additional 10.25 per cent of water. In one year 27 cc. of carbon dioxid were produced, giving a concentration of carbon dioxid of nearly 12 per cent. The deterioration in vitality was from 84 to 0 per cent, as compared with the control.

Lettuce.—Of air-dried lettuce seed, 10 grams were allowed to absorb an additional 8.87 per cent of water. During the experiment 19.5 cc. of carbon dioxid were formed, an equivalent of approximately 10 per cent of the original volume of the inclosed air. These seeds were all killed. The control sample germinated 94 per cent.

Onion.—Of air-dried onion seed, 10 grams were allowed to absorb an additional 10.11 per cent of water. The seed gave off 26.5 cc. of carbon dioxid during the experiment and deteriorated in vitality from 97 to 0 per cent.

A bottle containing 4 cc. of water was also sealed at the same time and served as a check for the other analyses. A sample of air taken from this bottle gave the same results as the original air sample.

It is a matter of much regret that no analyses could be made of the air from the bottles which contained the check samples. These bottles contained the same weight of air-dried seeds as was used for the experiments. Unfortunately the seals on these bottles had become dry and admitted of an exchange of gases, so that the results were not reliable.

Another series of experiments consisted in keeping onion seeds in sealed bottles for 1 year and 13 days, with the following results:

(a) Fifty grams of air-dried seed were sealed, in air, in a bottle of 500 cc. capacity. There was an increase in the weight of the seeds of 0.1091 gram—slightly more than 0.2 per cent. An analysis of the inclosed gas gave:

	Per cent.
Oxygen	. 12.27
Nitrogen	. 85.87
Carbon dioxid	1.86

(b) Fifty grams of air-dried seed were sealed, in air, in a 500 cc. bottle, with 4 cc. of water in a small test tube at the bottom of the bottle. Nearly all of the water was absorbed by the seeds, there being an increase in weight of 3.6475 grams, or 7.3 per cent. The composition of the inclosed air was:

	Pe	r cent.
Oxygen		None
Nitrogen		
Carbon dioxid		

The oxygen had all been consumed and the seeds were all dead.

<sup>(</sup>c) Fifty grams of onion seed were sealed in a 500 cc. bottle, in a

mixture of illuminating gas and air. The increase in weight was only 0.04 per cent. An analysis of the inclosed gas was as follows:

I	er cent.
Oxygen	3, 23
Carbon dioxid	1.21
Methane and nitrogen	95.96

(d) Another 50-gram sample of onion seed, belonging to a different series, was sealed in a bottle of 300 cc. capacity, and showed the following composition of the inclosed air:

Pe	r cent.
Oxygen	8, 02
Nitrogen	85. 17
Carbon dioxid	6.81

In only one case was there any deterioration in vitality, namely, where the large quantity of moisture was present. The other samples germinated normally. The seed kept in the illuminating gas germinated even better than the control.

In all of the bottles there was a marked decrease in pressure, showing that the volume of oxygen absorbed was much greater than the volume of the carbon dioxid given off.

During respiration certain chemical changes must be taking place which exert a marked influence on the vitality of seeds. What these changes are is a question yet to be solved. The protoplasts of the individual cells gradually but surely become disorganized. C. De Candolle" takes the view, in discussing the experiments of Van Tieghem and Bonnier, that during respiration life is simply subdued. But the period of subdued activity, he says, is comparatively short, for respiration soon ceases and life becomes wholly latent. As a result of his own experiments in storing seeds at low temperatures he concludes that seeds cease to respire and become completely inert; in which case they can suffer any degree of reduction in temperature without being killed. The killing of the seeds experimented with (lobelia) he attributes to the fact that the protoplasm had not become inert, but simply subdued, and the seeds were thus affected by the low temperature.

As a result of later experiments C. De Candolle, in keeping some seeds under mercury to exclude air, concludes that "seeds can continue to subsist in a condition of complete vital inertia, from which they recover whenever the conditions of the surrounding medium permits their 'energids,' or living masses of their cells, to respire and assimilate." He compares the protoplasm in latent life to an explosive mixture, having the faculty of reviving whenever the conditions are favorable. This comparison seems rather an unfortunate one; yet, within a certain measure it is probably true.

a Revue Scientifique, ser. 4, 4: 321-326, 1895.

<sup>&</sup>lt;sup>b</sup> Pop. Sci. Monthly, **51**: 106-111, 1897.

It is now quite generally accepted that respiration is not absolutely necessary for the maintenance of seed life, notwithstanding the fact that Gray contended that seeds would die of suffocation if air were excluded. The experiments of Giglioli in keeping seeds of Medicago sativa immersed in various liquids for approximately sixteen years, after which many responded to germination tests, has done much toward demonstrating the fact that seeds can live for a considerable time in conditions prohibiting respiration.

Kochs<sup>c</sup> succeeded in keeping seeds for many months in the vacuum of a Geissler tube without being able to detect the presence of any carbon dioxid, and consequently he concluded that there was no gas

given off by intramolecular respiration.

Romanes d kept various seeds in vacuum in glass tubes for 15 months and the seeds were not killed. However, his vitality tests can not be considered as entirely satisfactory. In the first place, the number of seeds used (ten) was too small; secondly, the variations in the results, even in the controls, indicate that the samples were not of very good quality.

In the experiments of the writer cabbage and onion seed were kept in a vacuum over sulphuric acid for 182 days. During this time all of the free water had been extracted from the seed. When again connected with a vacuum gauge the dial showed that there was not the slightest change in pressure, and that consequently no evolution of gases had taken place. The cabbage germinated 75 per cent and the onion 73 per cent as compared with 81 and 74 per cent, respectively, for the controls.

The results of the various experiments above given demonstrate quite fully that the vitality of seeds, as we commonly know them, is not interfered with if they are kept in conditions prohibiting respiration. Brown and Escombe hold that all chemical action ceases at temperatures of liquid air. They accordingly conclude that "any considerable internal chemical changes in the protoplasts are rendered impossible at temperatures of -180 to -190 C., and that we must consequently regard the protoplasm in resting seeds as existing in an absolutely inert state, devoid of any trace of metabolic activity and yet conserving the potentiality of life \* \* \* And since at such low temperatures metabolic activity is inconceivable an immortality of the individual protoplasts is conceivable providing that the low temperatures be maintained."

a Amer. Jour. of Sci., 3d series, 24: 297, 1882.

<sup>&</sup>lt;sup>b</sup> Nature, **52**: 544, 1895.

<sup>c Biol. Centrbl., 10: 673-686, 1890.
d Proc. Roy. Soc., 54: 335-337, 1893.</sup> 

e Ibid., 62: 160-165, 1897-98.

Giglioli arrived at practically the same conclusions when he said:

It is a common notion that life, or capacity for life, is always connected with continuous chemical and physical change \* \* \* The very existence of living matter is supposed to imply change. There is now reason for believing that living matter may exist, in a completely passive state, without any chemical change whatever, and may therefore maintain its special properties for an indefinite time, as is the case with mineral and all lifeless matter. Chemical change in living matter means active life, the wear and tear of which necessarily leads to death. Latent life, when completely passive in a chemical sense, ought to be life without death.

But even though ordinary respiratory exchanges are not necessary for the maintenance of vitality, and granting that intramolecular respiration does not occur in the resting protoplasts, there is no experimental evidence pointing to the fact that all chemical action ceases, although some writers, as has already been shown, maintain the view that living matter may exist in a completely passive state. If "completely passive" meant devoid of respiratory activities none would dare dissent; but that seeds are entirely quiescent under any known conditions has not been proved. To conceive of all activity ceasing within the seed under certain conditions, and that with such cessation of activity an immortality of the seed is possible, i. e., if such conditions continue to exist, is, from our present knowledge of the chemistry and behavior of the living cell, impossible. In Giglioli's experiments respiration was undoubtedly prevented, and, according to his own conclusions, vitality should have been preserved, for he says "in the absence of any chemical change the special properties may be maintained indefinitely." But, in his own experiments, the special properties were not maintained, for all of the seeds with which he experimented deteriorated very much, and many died. Granting that those which suffered the greatest loss in vitality were injured by the presence of the particular gas or liquid used there remain no means of accounting for the deterioration in those giving the highest percentages of germination. His experiments were made for the most part with Medicago sativa, which, under ordinary conditions of storage, is especially long lived. Samek b has shown that seed of Medicago sativa 11 years old was capable of germinating 54 per cent. Giglioli succeeded in getting a germination of only 56,56 per cent after a little more than 16 years in hydrogen, and 84.20 per cent when they had been kept in carbon monoxid. Jodin kept peas immersed in mercury for 44 years and they germinated 80 per cent. After 10 years the vitality had fallen to 44 per cent. Nobbe obtained a germination of 33 per cent in peas 10 years old which had been stored under normal conditions. Likewise the experiments of Brown and Escombe do not justify the

a Nature, 52: 544-545, 1895.

<sup>&</sup>lt;sup>b</sup> Tirol. landw. Blätter, 13: 161-162, 1894.

c Ann. Agron., 23: 433-471, 1897.

conclusions which they have drawn. It is now definitely known that all chemical actions do not cease at the temperature of liquid air. Thus it can not be granted that the protoplasm becomes inert as a result of the reduction in temperature. Maguenne a more nearly expressed the true conditions applicable to low temperatures when he wrote that with dessication, at low temperatures, seeds are transformed from a condition of diminished activity into a state of suspended life. But there are still other factors to be considered. The vegetative functions may cease, metabolic processes may be at a standstill, intramolecular respiration need not exist, yet vitality is not, nor ever has been, preserved; sooner or later life becomes extinct. What does this signify? The gradual process of devitalization means chemical change, and chemical change means activity within the cells. We must not forget the great complexity of the composition of the protoplasmic bodies which go to make up a seed. The chemistry of the living cell is still surrounded by many difficulties and is likewise filled with many surprises, and before the question of the vitality of seeds can be understood a more comprehensive knowledge of both the functions and composition of the cell contents is necessary.

It is well known that all organic compounds are made up of a very few elementary substances, but the numerous and obscure ways in which they are put together furnish questions of the greatest perplexity. Substances having the same elements may differ widely as to their properties. Moreover, isomeric substances—i. e., those having the same elements in the same proportions, giving an equivalent molecular weight—are usually very different in their chemical reactions and physiological functions. As yet this intramolecular atomic rearrangement is but vaguely understood, and the writer ventures to suggest that with a more comprehensive knowledge of the chemistry of the living cell some such chemical activity will be discovered. With these discoveries will come, perhaps, an understanding of the devitalization of seeds, and with it the theory of the immortality of seeds will vanish.

## SUMMARY.

(1) Seeds, like other living organisms, respire when subjected to normal conditions of storage.

(2) Respiration means a transformation of energy, and consequently a premature death of the seed.

(3) Within certain limits respiration is directly proportional to the amount of water present in the seeds and to the temperature at which they are stored.

(4) By decreasing the water content of seeds respiration is reduced and vitality greatly prolonged.

- (5) In most seeds the quantity of oxygen absorbed greatly exceeds the quantity of carbon dioxid evolved.
  - (6) Respiration is nearly as active in the dark as in the light.
- (7) Respiration apparently is not necessary for the maintenance of seed life.
- (8) A cessation of respiration does not mean a cessation of chemical activities.

# ENZYMES IN SEEDS AND THE PART THEY PLAY IN THE PRESERVATION OF VITALITY.

During the past decade the so-called unorganized ferments have taken an important place among the subjects of biological research. Our knowledge of their wide distribution has increased many fold. The part they play in both anabolism and catabolism has furnished us many surprises, but with all of the work that has been done our knowledge of these most complex compounds is very limited.

The part that enzymes play in the processes of germination is of the utmost importance. It is now quite well understood that they are developed as germination progresses. They act on the most complex reserve food products, converting them into simpler substances that can be more readily utilized by the growing seedling.

However, even in this connection there is a great diversity of opinion, especially as to their distribution and enzymic action within the endosperm itself. Puriewitsch," Grüss, hand Hansteen are cited by Brown and Escombe as holding the view that the amyliferous cells of the endosperm of the grasses can digest their reserve materials independently of any action of the embryo—i. e., the starch-bearing cells are living cells and secrete enzymes in the grasses as well as in the cotyledonous cells of Lupinus, Phaseolus, and Ricinus. In 1890, Brown and Morris did not find such to be the case; but the results of Puriewitsch, Grüss, and Hansteen led to a duplication of the experiments by Brown and Escombe in 1898. At this time they demonstrated that the amyliferous cells play no part in the chemical changes which take place during the process of germination, but on the contrary that the enzymic action in the endosperm of the grasses is confined to the aleuron layer.

But the purpose of the present paper is not to consider the localization of the particular enzyme, and much less the action of enzymes during germination. At this time quite another question is to be

a Pringsheims Jahrb., 31: 1, 1897.

<sup>&</sup>lt;sup>b</sup> Landw. Jahrbücher, 1896, p. 385.

c Flora, 79: 419, 1894.

d Proc. Roy. Soc., 63: 3-25, 1898.

e Jour. Chem. Soc., London, 57: 458-528, 1890.

considered, viz, In what way do enzymes function in the preservation of vitality?

Maquenne points to the view that the vitality of seeds is dependent on the stability of the particular ferment present. He attributes the prolongation of vitality in seeds that are kept dry to the better preservation of the enzymes. This view has been largely strengthened as a result of the investigations made by Thompson, Waugh, Sharpe, and others, in which they have shown that the artificial use of enzymes may greatly increase the percentage of germination in some old seeds. By the use of diastase the percentage of germination of 12-year-old tomato seed has been increased more than 600 per cent.

If the suggestions made by Maquenne were true in every sense, then dead seeds should be awakened into activity by artificially supplying the necessary enzymes; but this can not be, or never has been, accomplished. True, many experiments have been recorded in which a greater percentage of seed has been induced to germinate by the judicious use of commercial enzymes than by the ordinary methods of germination; but this treatment is applicable only where the vital energy is simply at a low ebb and does not in any way affect dead seeds. The experiments of the writer with naked radicles from the embryos of living and dead beans have shown the presence of enzymes in both. The carefully excised radicles were ground and macerated in water for one hour. The filtrate was then added to dilute solutions of starch paste. The solutions from the living embryos gave rise to an energetic hydrolytic action. In all cases hydrolysis was sufficiently advanced to give a clear reaction with Fehling's solution. The solutions extracted from the radicles from the dead beans also gave reactions sufficiently clear to indicate that there was still some ferment present.e

However, the hydrolysis was scarcely more than begun, giving only a brown color with iodin, but not reacting with Fehling's solution. Results of a similar character were obtained from portions of the seed

aAnn. Agron. 26: 321-332, 1900; Compt. Rend., 134: 1243-1246, 1902.

<sup>&</sup>lt;sup>b</sup> Gartenflora, **45**: 344, 1896.

Ann. Report, Vt. Agr. Exp. Sta., 1896-97, and Science, N. S., 6: 950-952, 1897.
 Thirteenth Annual Report, Mass. Hatch Exp. Sta., Jan., 1901, pp. 74-83.

eThis was a sample of "Valentine" beans grown in 1897. The same year they tested 97.3 per cent. In March, 1898, the same sample tested 87 per cent. At this time they were sent to Orlando, Fla., where they remained until May 8, 1899, approximately fourteen months. The beans were then returned and numerous germination tests were made at irregular intervals, but in no case was there any indication of vitality. Several samples were also treated with "Taka" diastase (solutions varying in strength from 2 to 10 per cent), but none was stimulated into germination. The radicles were tested for enzymes in the spring of 1902, nearly three years after the beans first failed to germinate, at which time they were nearly 6 years old.

taken from the point of union of the axis and the cotyledons. These possessed stronger hydrolytic powers, the preparations from the living and dead beans each giving clear reactions with Fehling's solution. A third series of tests was made by stopping the germination of beans when the radicles were from 1 to 1.5 cm. long. These were then kept quite dry for nearly seven months, after which the dessicated radicles were broken off and macerated like the above. This solution was then allowed to act on starch paste, and the transformations were almost as rapid and complete as when a 1 per cent solution of commercial "Taka" diastase was used.

These results lead one to believe that the loss of vitality in seeds is not due to the disorganization of the enzymes present. There is something more fundamental and probably more complex to which we must look for this life-giving principle. True, as Maquenne has suggested, there is a close relationship between the loss of vitality in seeds and the decomposition of enzymes.

In order to determine what such a relationship might signify, the following series of experiments were made:

Beans, peas, cabbage, lettuce, onion, phlox, and pansy seed, with definite quantities of good commercial "Taka" diastase, were put up in bottles of 120 cc. capacity, as follows:

- (1) In bottle closed with cork stopper.
- (2) In bottle closed with cork stopper and paraffined.
- (3) 0.5 cc. of water in the bottle with the seeds and the diastase, the bottle sealed with paraffin.
- (4) 1 cc. of water in the bottle with the seeds and the diastase, the bottle sealed with paraffin.
- (5) 2 cc. of water in the bottle with the seeds and the diastase, the bottle sealed with paraffin.
- (6) 3 cc. of water in the bottle with the seeds and the diastase, the bottle sealed with paraffin.
- (7) 4 cc. of water in the bottle with the seeds and the diastase, the bottle sealed with paraffin.

The water in each case was carefully added on small strips of filter paper and never were the seeds or the diastase wet, only becoming gradually moist as the water was absorbed.

These different preparations, each containing one of each of the samples of seeds and a definite quantity of the dry powdered diastase, were then maintained at the temperature of the laboratory for a period of 85 days. At the end of that time the vitality of the seeds was determined and simultaneously the hydrolytic power of the diastase was ascertained. The results of the germination tests are given in Table XXIX. The effect of the increased quantity of moisture on the diastase is given in the discussion following the table.

Table XXIX.—Loss in vitality of seeds with varying degrees of moisture when kept at ordinary room temperature.

[Duration of experiment, 85 days.]

Labor-		Amount			Percenta	ige of geri	nination.		_
atory num- ber.	i- of sample.	of water added.	Beans.	Peas.	Cabbage.	Onion.	Phlox.	Pansy.	Average of all samples.
		cc.							
	Control a	None	96, 0	90,0	91.5	95, 0	41.25	46, 0	76.6
1547	Corked	None	98.0	96.0	91.0	92.5	52.0	32, 0	76.9
1548	Paraffined	None	96.0	92.0	91.5	93.0	39.5 .	31.0	73.8
1549	do	0.5	96.0	92.0	89.0	88.8	28.5	25, 5	69.9
1550	do	1.0	96.0	88.0	89.0	64.0	12, 5	18.0	61.2
1551	do	2.0	96.0	86.0	78.0	13.0	. 5	2.5	46.0
1552	do	3.0	94.0	94.0	65, 0	2.5	. 5	. 5	46.1
1553	do	4.0	90.0	81.6	54.5	.0	. 0	.0	37.6

lpha The samples prepared, excepting the control, were in bottles of 120 cc. capacity.

The above table shows that there was a gradual deterioration in vitality as the quantity of water was increased. All stages of injury were manifested, but it is not necessary to enter into a discussion of the table at this time, inasmuch as similar tabulations, showing the injurious effects of varying quantities of moisure on the seeds, have already been given on page 38. This table is inserted here in order that a comparison can be made with the decomposition of the commercial diastase used and the loss in vitality of the seeds.

For a determination of the diastasic activity various quantities of 1 per cent "Taka" diastase solutions were allowed to act on definite quantities of a 1 per cent solution of starch paste, the whole being maintained at a temperature of from 45° to 48° C. Ten cubic centimeters of the starch solution were taken for each determination, and the amount of the diastase solution varied from one-half to 1, 2, 3, and 5 ce. In the control sample, consisting of diastase from the original bottle as it was kept in the laboratory, 2 cc. of the 1 per cent solution were sufficient to cause a complete hydrolysis of the 10 cc. of 1 per cent starch solution. In Nos. 1547, 1548, and 1549 the samples from the control bottle, the paraffined bottle, and the paraffined bottle containing 0.5 cc. of water, respectively, 3 cc. of the diastase solution were necessary for a complete hydrolysis. In Nos. 1550, 1551, and 1552—that is, the samples from the bottles which contained 1, 2, and 3 cc. of water, respectively the diastase was very much injured as a result of the increased quantity of water in the bottle and 5 cc. of the diastase solution were required to hydrolyze the 10 cc. of the 1 per cent starch paste. No. 1553—the sample from the bottle which contained the 4 cc. of water showed that the diastase had been almost completely disorganized, inasmuch as the greatest quantity used (5 cc. of the 1 per cent diastase solution) was only sufficient to cause a slight hydrolytic action. When

tested with iodine there was still a deep, purplish-blue color. In this last case the average percentage of germination had decreased to 37.6 per cent, as compared with 76.6 per cent for the control samples. Moreover, in the latter case, the onion, phlox, and pansy seeds were killed.

These results show that there is a remarkable uniformity between the loss in vitality of seeds and the loss in the enzymic action of the "Taka" diastase under similar conditions, but it does not furnish conclusive evidence that the loss in vitality of the seeds is in any way governed by the particular enzymes present. In fact, the evidence at hand better substantiates the opposite view. In the first place dead seeds may still contain active ferments. Secondly, the prolonged subjection of seeds to the action of ether and chloroform is generally accompanied by a premature death, and if the seeds are moist the loss in vitality is much more marked. On the other hand, it is generally accepted that either of these gases exerts no injurious effect on the hydrolytic action of the various ferments. Townsend a has shown that the action of diastase on starch paste is even more energetic in the presence than in the absence of ether, but in germination ether usually has a retarding influence. In some cases, however, growth is stimulated by the use of ether.

In the third place enzymes can not be the chief factors controlling the vitality of a seed, because the more sensitive growing point of the radicle suffers injury much in advance of the other portions of the seed. Not infrequently in making germination tests do we find that the growing tip of the embryo is dead, while other portions of the seed may still be living and capable of carrying on all normal metabolic processes. The bean is one of the best examples for demonstrating this fact. Here the radicle may be entirely dead, yet the cotyledons may still be able to make some growth; but in all seeds where the growing tip is dead the remaining portion of the radicle may be living, in which case adventitious roots may be formed and growth may continue for a considerable time, though very rarely will a healthy seedling be developed. It thus seems quite clear that the real vital elements are closely associated with the growing point, and when this portion of the embryo is once dead the vital energy in the other parts of the seed is not of such a nature as to enable growth to continue for any length of time. Even though the reserve food products are digested they can not be assimilated by the growing radicle, which should be the case were enzymes the chief elements to which the preservation of vitality is attributed.

Enzymes play an important part in the vitality of seeds, and are undoubtedly necessary for the normal development of a seedling, but the points above given show that the life of a seed is not entirely dependent on the stability of the particular ferment or ferments present. There is something more remote, possibly of a simpler but probably of a more complex composition, to which we must attribute the awakening of the metabolic processes. Reference is not made here to the zymogenic substances which develop into the particular ferment, for what has been said of the latter applies equally well to the former. If the zymogens were perfectly preserved the resulting ferments would be developed normally and germination would continue in the usual manner.

In conclusion, it may well be emphasized that no single element or compound can be isolated as the sole source of vitality in seeds. There must be a combination of factors, each of which plays an important rôle in the preservation of vitality. The destruction of any one of these factors may upset the principles governing the life of a seed, and consequently cause a premature death.

It is quite probable that the nucleus is one of the most important organs governing vitality, for unless it continues to function no other growth can take place. Other parts of the cell, however, may be of equal importance. At all events all hope of future gain must come from more critical studies of the cell contents to know their chemical composition and possible reactions. A correct solution of these perplexing questions is nothing less than a determination of the fundamental principles of life. What will be the ultimate results no one is prepared to say.

#### SUMMARY.

(1) A seed is a living organism, and must be dealt with as such if good results are expected when put under favorable conditions for germination.

(2) The first factors determining the vitality of a seed are maturity, weather conditions at the time of harvesting, and methods of harvest-

ing and curing.

(3) Immature seeds sown soon after gathering usually germinate readily, but if stored they soon lose their vitality. On the other hand, well-matured seeds, harvested under favorable conditions, are comparatively long lived when properly handled.

(4) Seed harvested in damp, rainy weather is much weaker in vitality than seed harvested under more favorable conditions. Likewise,

seed once injured will never regain its full vigor.

(5) The curing of the various seeds is of the utmost importance, and great care should be taken to prevent excessive heating, otherwise the vitality will be greatly lowered.

(6) The life period of any species of seed, granting that it has been thoroughly matured and properly harvested and cured, is largely dependent on environment.

(7) The average life of seeds, as of plants, varies greatly with different families, genera, or species, but there is no relation between the longevity of plants and the viable period of the seeds they bear. The seeds of some plants lose their vitality in a few weeks or months, while others remain viable for a number of years.

(8) With special precautions and treatment there is no question that the life of seeds may be greatly prolonged beyond that which we know at present, though never for centuries, as is frequently stated. Cases so reported can not be taken as evidence of the longevity of seeds.

(9) It is known that seeds retain their vitality much better in some sections of the country than in others. The part which climatic influences play in the vitality of seeds is of much more importance than is

generally supposed.

(10) Experiments have shown that *moisture* is the chief factor in determining the longevity of seeds as they are commercially handled. Seeds stored in dry climates retain their vitality much better than when stored in places having a humid atmosphere.

(11) The deleterious action of moisture is greatly augmented if the temperature be increased. Not infrequently is vitality destroyed within a few weeks or months when the seeds are stored in warm, moist climates. If stored in a dry climate, the question of temperature within the normal range is of little moment.

(12) The storage room for seeds as they are ordinarily handled should always be *dry*. If seeds could be kept dry and at the same time cool, the conditions would be almost ideal for the preservation of vitality; but the difficulties to be overcome in order to secure a dry and cool storage room render this method impracticable.

(13) The most feasible method for keeping seeds dry and thus insuring strong vitality is to store them in well ventilated rooms kept dry by artificial heat. This method of treatment requires that the seeds be well cured and well dried before storing.

(14) If seeds are not well dried vitality is best preserved at temperatures just above freezing, provided that the temperature is maintained uniformly.

(15) In no case must the temperature of the storage house be increased unless the seed is amply ventilated so that the moisture liberated from the seed can be carried off readily by the currents of warm air. If this precaution is not taken the increased humidity of the air confined between the seeds will cause a marked injury. For this same reason seeds kept at low temperatures during the winter will deteriorate in the warm weather of spring, especially if they contain much moisture.

(16) Most seeds, if first carefully dried, can withstand long exposures to a temperature of 37 °C. (98.6 °F.) without injury, but long exposures to a temperature of from 39 to 40 °C. (102.2 °to 104 °F.)

will cause premature death. If the seeds are kept in a moist atmosphere a temperature of even 30° C. (86° F.) will soon cause a marked injury.

- (17) Seeds can endure any degree of drying without injury; that is, by drying in a vacuum over sulphuric acid. It is believed that such a reduction in the water content is necessary if vitality is to be preserved for a long period of years. However, with such treatment the seed coats become very firm, and there usually follows a retardation in germination as a result of the inability of the seeds to absorb water rapidly enough to bring about the necessary physical and chemical transformations for the earlier stages of germination.
- (18) Seeds that are to be sent to countries having moist climates should be put up in air-tight packages. Experiments have shown that by the judicious use of bottles and paraffined packages seeds can be preserved practically as well in one climate as in another.
- (19) It is of the utmost importance that the seeds be dry before being sealed in bottles or paraffined packages. A drying of ten days at a temperature of from 30° to 35° C. (86° to 95° F.) will usually be sufficient. However, a better method to follow is to dry until no more moisture is given off at a temperature equivalent to the maximum of the region in which the seeds are to be distributed. If this is not done, the subsequent increase in temperature will liberate an additional quantity of moisture, which being confined in the package will leave the seeds in a humid atmosphere and a rapid deterioration in vitality will follow.
- (20) Experiments in storing seeds in open and sealed bottles and in packages with definite quantities of moisture and at various known temperatures have shown a very close relationship between the loss in vitality and the increase in water content, the deterioration likewise increasing with the temperature.
- (21) Of a series of experiments the average loss in vitality of seeds kept in envelopes in a "dry room" was 21.19 per cent, "trade conditions" 36.63 per cent, "basement" 42.28 per cent, while the loss in the case of seeds stored in bottles was only 8.08, 3.92, and 4.51 per cent, respectively. (See Table XXV.)
- (22) Seeds under ordinary conditions of storage respire quite freely, and respiration is much more rapid if much moisture is present. Within certain limits respiration is directly proportional to the amount of moisture present in the seed and inversely proportional to the duration of vitality.
- (23) Respiration is not necessary to the life of seeds, as they can be kept in conditions unfavorable for respiratory activity and still retain their vitality even better than under normal conditions of storage. Even though respiration be entirely prevented seeds will continue to deteriorate, and sooner or later lose their vitality.

- (24) The continued deterioration in the vitality of a seed after respiration has ceased indicates a chemical activity within the cells, giving rise to a transformation of energy which sooner or later leads to the death of the seed.
- (25) Respiration is almost as active in the dark as in the light, provided that the temperature and humidity remain the same.
- (26) Ferments and seeds lose all power of activity under similar conditions of moisture, and the former are undoubtedly of the utmost importance in metabolic activity, but the evidence at hand goes to show that the life of a seed is not dependent on the preservation of the particular ferment involved or on the zymogenic substances giving rise to the enzyme.
- (27) The life of a seed is undoubtedly dependent on many factors, but the one important factor governing the longevity of good seed is dryness.

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